Predisposing factors, the role of underlying thyroid dysfunction, and tests for early detection.

1990 Update on Autoimmune Diseases

BY W. JEAN DODDS, D.V.M.

Pure-bred dogs fanciers have contacted the Wadsworth Center for Laboratories and Research (of the N.Y. State Department of Health) again recently requesting information about the increasing frequency of autoimmune problems in their breeds. This article reviews the factors that predispose to autoimmune diseases with special emphasis on the role of underlying thyroid dysfunction and the early detection of susceptible individuals.

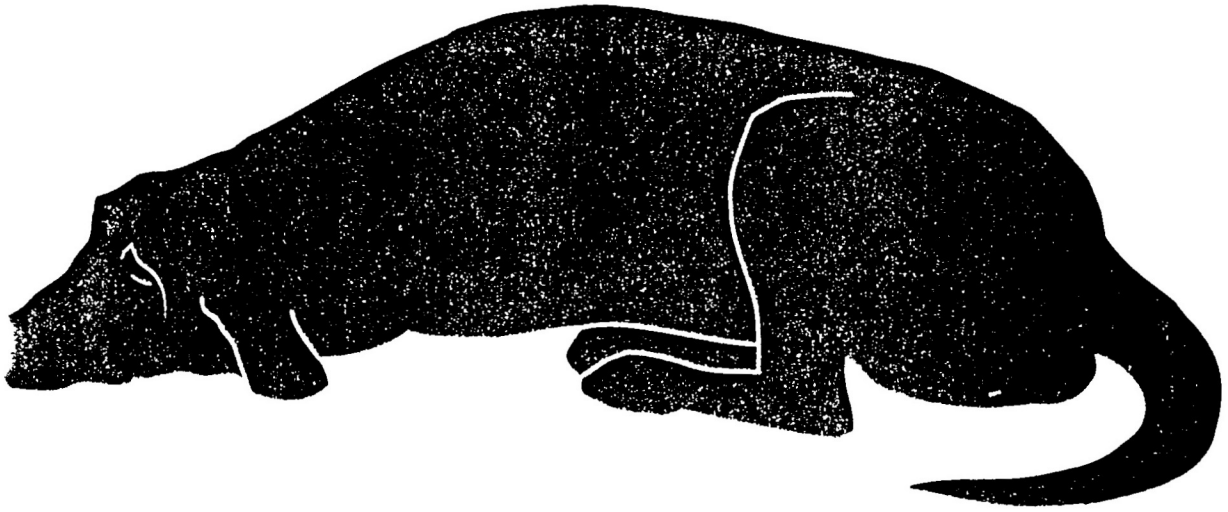
Predisposing Factors

In recent years the prevalence of autoimmune (immune-mediated) diseases has been increasing rapidly in man and animals. This has been attributed to such factors as genetic and sex predisposition, nutritional influences, concomitant viral infections or frequent use of modified live virus vaccines, and pituitary-thyroid axis imbalances. (See sidebar, "Factors Associated With Autoimmune Disease.") Specifically with respect to the number of individuals genetically susceptible to autoimmune thyroid disease, the process is believed to start around puberty as thyroiditis, and progress gradually through mid-life and old age to become clinically expressed hypothyroidism once thyroid reserve has been depleted. During this process the individual becomes more susceptible to immune-mediated diseases affecting other target tissues and organs (polyglandular autoimmune syndrome), especially the bone marrow, liver, adrenal gland, pancreas, skin, kidney, joints, bowel and central nervous system.

There are four primary causes of autoimmune disease and autoimmune endocrine (for example, thyroid) disease in particular: a) genetic predisposition; b) viral infection or exposure; c) hormonal influences especially of sex hormones; and d) stress. In humans with a parallel form of thyroid disease (Hashimoto's thyroiditis or lymphocytic thyroiditis), they have identified a specific profile of the major histocompatability complex, HLA-DR, which conveys genetic susceptibility to the disease. Furthermore, in May 1989 a research team in England discovered a

Dr. Dodds is director of the Comparative Hematology Laboratory, N.Y. State Department of Health in Albany. She is a breeder/handler of Pointers, English Setters and Vizslas.

SEX ... GENETIC HISTORY
INCREASING FREQUENCY
DRUG REACTIONS ... STRE
HORMONAL IRREGULARI



novel retrovirus, distinct from but related to the HIV virus that transmits AIDS, that is associated with human thyroiditis. The bottom line is that viruses capable of inducing immune dysregulation in genetically susceptible dogs can initiate autoimmune thyroid disease under the appropriate environmental conditions (e.g., hormonal imbalance, stress, drug or toxin exposure, dietary factors, etc.). There is every reason to believe that the canine disease has similar causation. In the past several months, three veterinary groups have demonstrated reverse transcriptase activity (a result of retrovirus exposure) in canine lymphoma/leukemia cell lines. Dogs and especially closely related linebred/inbred purebreds have the genetic makeup that conveys susceptibility, the viral elements are there, and reproductive hormonal and stress influences are prevalent when one breeds for any type of

an early manifestation of hypothyroidism. This includes easy bruising and thrombocytopenia, abnormal platelet function which returns to normal after treatment with thyroid hormone, and acquired vWD similar to the common Type I form of this disorder (low vWF:Ag). *It is generally impossible to distinguish between the inherited and acquired types of vWD in an individual patient with currently available techniques.*

Circulating antithyroid antibodies are often present in dogs with autoimmune thyroid disease (called thyroiditis or Hashimoto's thyroiditis) several years before the thyroid disease becomes clinically apparent. Thus animals with thyroid dysfunction can have fluctuating levels of vWF and, when placed on thyroid supplementation, levels can increase to within normal limits, which could preclude the accurate diagnosis of their genetic status for vWD (i.e., carriers of vWD might test as normal when on thyroid medication). Because of this apparent exacerbation of bleeding tendencies in dogs with vWD with hypothyroidism, breeders and veterinarians need to be aware of the increased risk and be more cautious about breeding or performing surgery on dogs with both problems.

In humans, antithyroid antibodies can be transferred from mothers with Hashimoto's thyroiditis to their infants in breast milk. Neonatal hypothyroidism can occur and is picked up on the routine newborn screening programs run by state and local public health agencies. Whether a parallel transfer of maternal antithyroid antibody occurs via the colostral milk to newborn puppies is unknown but is likely. What effect, if any, the transfer of these antibodies could have on the development and health of puppies is also unknown. Both humoral and cell-mediated immunity could be affected, especially when growth hormone and thyroxine modulate the thymus and thereby immune function. Perhaps the presence of maternal circulating antithyroid antibodies during the first several months of life could alter immune recognition or development sufficiently to render these puppies more susceptible to other immune-mediated stressors (viruses, vaccines, drugs, toxins, etc.). These antibodies could play a role in stimulating the active immunlogical process that leads to the development of immune thyroiditis.

The severely limited availability of blood products for clinical use in veterinary medicine has necessitated the development of alternate strategies for the treatment and management of bleeding disorders. In breeds in which both vWD and hypothyroidism occur relatively often, the administration of oral thyroid hormone has been effective in controlling bleeding episodes exacerbated by these two diseases. Clinical signs of bleeding are lessened or controlled within forty-eight hours after therapy is initiated. In man, L-thyroxine therapy has been shown to reduce levels of the circulating antibodies responsible for thyroiditis and hypothyroidism. Presumably this plays a role in the reversal of the bleeding tendency associated with thyroid disease. In dogs, the toenail bleeding time is dramatically shortened and in some cases may even be corrected to within normal limits (less than five minutes) from pretreatment values beyond twenty minutes. Thus, *thyroid supplementation alone may suffice to control bleeding in mild to moderate vWD*, a situation analogous to the use of desmopressin (DDAVP) or to improve vWF levels in normal dogs and dogs with vWD, although relatively high doses are required and the response is transient (one to three hours); refractoriness to this treatment can develop rapidly.

The standard dosage of L-thyroxine for treatment of vWD (enhances platelet stickiness [adhesion] and improves vWF activity) is 0.1 milligrams/10 pounds body weight given twice daily. The therapeutic response should be monitored with serial toenail or other bleeding times and periodically by thyroid

function tests: dosages of medication should be adjusted accordingly to maintain thyroid levels (total T4, free T4, total T3) within the upper half to third of the adult normal ranges.

VWF Production/Auto. Thy. Dis.

The endothelial cells lining blood vessels are the primary and essentially sole source of vWF production in the dog. Thyroid dysfunction from familial autoimmune thyroiditis is by far the most commonly recognized cause of acquired vWD in dogs. Production and/or secretion of vWF from endothelial cells is clearly altered by thyroiditis. The most likely cause of lower than expected vWF:Ag scores on dogs with parents known to be free of the vWD gene is therefore this thyroid disorder, whether or not it is expressed to date, because the pathologic process associated with autoimmune antithyroid antibody production can take several years to induce the end stage of clinical hypothyroidism. If you breed these dogs they will transmit susceptibility for the thyroid problems to some of their offspring.

Our cumulative data on many breeds affected by familial autoimmune thyroid disease indicate that reduction in vWF:Ag production associated with thyroiditis is becoming quite common. Since the clinical and laboratory features of this acquired form of vWD are indistinguishable from the congenital inherited form of vWD, and the inheritance and carrier detection program for vWD has not changed, the situation over the past decade, and especially in the last five years, is complicated by the rapid rise in the incidence of thyroid and other autoimmune diseases in affected breeds.

We believe it is very important for pure-bred dog fanciers to understand the influence autoimmune thyroid disease has on vWF levels and the expression of acquired vWD. This is a complex and important problem which impacts the survival and vigor of affected breeds, and is more difficult to control than inherited vWD which can

PURCHASE EXAMS

I purchased a Cocker Spaniel in 1983 and an English Cocker in 1984, both dogs to be trained for obedience and tracking. Last September, at the age of six, my Cocker Spaniel died of complications from the drugs used to treat her immune-mediated hemolytic anemia and thrombocytopenia. My three-year-old English Cocker developed immune-mediated thrombocytopenia (IMT) in March 1988. Since then, she has required constant medical attention and daily care to control the IMT and complications arising from the medications she must take. Because of the expense and emotional toll associated with these events, it is important that my next puppy be healthy at the time of purchase. In addition, the puppy must be physically able to perform in obedience, tracking, and the work of the breed (whether that be field work or herding).

Early kidney/liver disease, von Willebrand's disease, low-grade infections and many other health conditions are not detectable during the physical exam routinely done on a new puppy. Consequently, I have decided to supplement the customary physical exam with the following tests: CBC, chemistry panel, platelet count, and urinalysis (to look for evidence of infection or congenital disorders); von Willebrand's disease test on the puppy and both parents; thyroid profile; EKG; OFA-type Xray (without anesthesia) if the puppy is at least four months old; complete eye exam; heartworm test if the puppy is at least six months of age; and gait assessment.

While these tests do not guarantee a future free of health problems, they do provide more assurance than a physical exam alone that a puppy has no major health defects at the time of sale and is capable of being worked. (If I had performed these tests on my Cocker Spaniel and my English Cocker, I would have returned the Cocker Spaniel, who had von Willebrand's disease, and kept the English Cocker, who had no health problems prior to developing IMT.)

When measured against the expense involved in acquiring and training a promising performance dog and the often considerable expense of veterinary medical care, the cost of the above tests (approximately $100 minimum, not including examinations) is minimal. Test results are usually available within two weeks or less so that a puppy can be returned promptly if significant health problems are detected. Additional tests may be appropriate depending on health problems prevalent in specific breeds and the age and intended use of the dog. These tests are not intended to be a substitute for tests performed by breeders at their expense to ensure that their puppies are free of common genetic defects; nor are they designed to replace specific warranties against genetic defects offered by breeders.

In the equine world, the sale of any horse purchased for performance reasons is conditional upon the results of a purchase exam, which determines the health potential of the horse to perform a given task. I believe that use of the purchase exam in the canine world would benefit buyer and breeder alike. Both can be more confident that, at the time of sale, a puppy/older dog is healthy and free of defects which would decrease its usefulness. Increased testing would also provide breeders with comprehensive information about the health of their stock, including early warning of hereditary defects.—**Sandra A. Priest, D.V.M.**

The author, an English Cocker Spaniel owner, participates in obedience and tracking. She also works in the computer field.

be addressed relatively easily with an open, honest testing and planned breeding program.

Clinical Signs of Canine Hypothyroidism

Alterations in cellular metabolism
- -lethargy
- -mental dullness
- -exercise intolerance
- -weight gain
- -cold intolerance
- -mood swings or hyperexcitability
- -neurologic signs (including polyneuropathy and seizures)

Reproductive disorders
- -infertility of either sex
- -lack of libido
- -testicular atrophy
- -hypospermia
- -aspermia
- -prolonged interestrus interval
- -failure to have heat cycles
- -weak, dying or stillborn pups
- -enlarged mammary glands or lactation in non-pregnant bitches (pseudopregnancy)

Neuromuscular problems
- -weakness
- -stiffness
- -laryngeal paralysis
- -knuckling or dragging feet
- -muscle wasting

Dermatologic diseases
- -dry, scaly skin
- -coarse, dull coat
- -bilaterally symmetrical hair loss
- -"rat tail"
- -"puppy coat"
- -hyperpigmentation
- -seborrhea
- -pyoderma
- -myxedema

Cardiac abnormalities
- -bradycardia or slow heart rate
- -cardiac arrythmias
- -cardiomyopathy

Ocular diseases
- -corneal lipid deposits
- -corneal ulceration
- -uveitis
- -kerato-conjunctivitis sicca or "dry eye"

Gastrointestinal disorders
- -constipation
- -diarrhea
- -vomiting

Hematologic disorders
- -bone marrow failure
- -bleeding

—WJD

Thyroiditis Testing

About ninety percent of thyroid abnormalities in the dog have an inherited autoimmune basis. As mentioned earlier, this condition is linked to other immune-mediated problems such as AIHA and ITP; bone marrow failure; leukemia and lymphoma; SLE; seizure disorders; chronic infections; immunosuppressive viral infections like distemper, parvovirus and retrovirus diseases; chronic active hepatitis (liver disease); immune kidney and adrenal (Addison's) diseases; chronic allergic and immune skin and muscle disorders.

Thyroiditis is the immune-mediated process characterized by the presence of antithyroid antibodies in the blood or tissues. This condition usually progresses eventually to thyroid disease.

Antithyroid antibodies can be present in euthyroid (normal), hypothyroid, or hyperthyroid humans or animals, i.e., can be present while a dog is still testing normal for thyroid function, and can be doing things to other tissues such as bone marrow, fetuses, and other fast-growing cells. Part of the management of autoimmune disorders is to test for the presence of thyroid dysfunction and antithyroid antibodies, and to treat with thyroid hormones to inhibit the process. Therefore, we need a way to diagnose the presence of antibodies before the disease develops.

A typical thyroid test measures the total presence of the T4 hormone in the body. But an important measurement is the small fraction of the total T4 which is not bound in the body and is therefore available to the tissues. This "free" T4 is converted in the tissues to T3, which is important for intracellular function.

In the newer thyroid profiles available at specialized veterinary laboratories in the U.S. and Canada, they test for total 14, total T3, free T4, cholesterol, and free T3. Cholesterol should be measured from the same blood sample as the thyroid tests. In dogs with thyroid dysfunction, the higher the cholesterol level, the lower the level of free T4. An owner can obtain a printout listing all the test results for his or her dog, as well as the K value, which is a linear constant between free T4 and cholesterol.

With respect to K values, those which are above +1 and preferably above +5 are considered to be normal. Less than -4 indicates an animal with primary thyroid disease, even if there are no overt symptoms. Values between -4 and +1 are either early thyroid disease and/or a nonthyroidal illness (another illness which depresses thyroid levels). Thyroid deficiency even at early subclinical stages can cause irreversible damage in the body, and the value of

"Hypothyroidism is the most common canine endocrine disorder."

this more sensitive test is that problems can be detected before damage has become so severe that clear visible symptoms are present.

For a dog displaying symptoms of autoimmune disease, the K value can be helpful to indicate whether the dog has low, normal or high thyroid function, and if the autoimmune problems are likely to be responsive to thyroid hormone supplementation. If test results are normal, and the dog is nevertheless ill, proceed to have other diagnostic tests done, such as that for antithyroid antibodies (antithyroglobulin, anti T4 and anti T3 autoantibodies). (See "Bleeding And Autoimmune Testing Facilities" sidebar.)

(Note: The thyroid profiling outlined above differs from the thyroid stimulating hormone [TSH] response test often used to diagnose the clinical stages of hypothyroidism. This test measures thyroid reserve by assessing the ability of the thyroid gland to release T4 hormone upon stimulation with an injection of TSH. It is estimated that from fifty to seventy percent of thyroid follicular tissue has to be destroyed or damaged before hormonal reserves are depleted to the extent that overt hypothyroidism occurs. Thus, in the early stages of progressive thyroid disease [thyroiditis], the TSH response is usually normal and would be misleading [i.e., a false negative diagnosis]. For this reason, we do not rely on finding a positive TSH response test to diagnose thyroiditis.)

Thyroglobulin is a glycoprotein found in thyroid gland follicles. The metabolites of thyroid hormones, T3 and T4, are incorporated in the primary structure of thyroglobulin. Thyroglobulin is antigenic, and specific antibodies can form that are directed against several sites on the molecule. In humans the presence of antithyroglobulin antibodies (ATA) is most often associated with Hashimoto's thyroiditis, the immune-mediated inflammation of the thyroid gland that progressively destroys functional thyroid tissue. The outcome of this process is hypothyroidism, and lifelong treatment with thyroid hormone is required. Hashimoto's thyroiditis is common; and cases tend to cluster among families and certain ethnic groups.

Hypothyroidism is the most common canine endocrine disorder. If the thyroid gland is biopsied, changes characteristic of Hashimoto's thyroditis are seen in most cases. Assays that measure ATA have been adapted for dogs. In two independent studies, about fifty to eighty percent of the clinically hypothyroid dogs tested had significant

Bleeding And Autoimmune Testing Facilities

The Wadsworth Center for Laboratories and Research will analyze blood samples from your dogs for bleeding disorders associated with autoimmune diseases. Have your veterinarian submit samples from your animals, indicating whether they have any type of health problem or family history of liver and/or thyroid disease. If your dogs are completely healthy at the time of testing, please also indicate this fact so that those receiving samples can avoid assumptions that may be incorrect.

Samples should be sent to Veterinary Hemotology Laboratory, Wadsworth Center for Laboratories and Research, New York State Department of Health, P.O. Box 509, Albany, NY 12201-0509, (518) 869-4507.

To locate the nearest veterinary testing laboratory that provides complete thyroid profiling, contact your veterinarian or regional veterinary teaching hospital. Several university and commercial veterinary testing laboratories in the U.S. and Canada now offer this service.

Michigan State University has recently introduced antithyroid hormone antibody (anti T4 and anti T3) testing as part of their complete thyroid panel. This is an important step towards more effective thyroid testing. Contact the Endocrinology Section, Animal Health Diagnostic Laboratory, P. O. Box 30076, Lansing, MI 48909, (517) 353-0621.

The College of Veterinary Medicine, University of Florida, Gainesville, FL 32610, (904) 392-4746, offers clinical antithyroglobulin antibody testing, and the Wadsworth Center for Laboratories and Research provides this test on a clinical research basis only.—**WJD**

levels of ATA. Positive ATA titers were also found in some dogs with nonthyroidal endocrine disease, and in some euthyroid relatives of hypothyroid dogs. At present, the prevalence of ATA in the various breeds affected with hypothyroidism, the value of ATA as a predictor of impending thyroid failure or a marker of familial hypothyroidism, and the relation of ATA to other immune-mediated disorders has not been determined.

Preliminary studies of several affected dog families show promise in supporting the usefulness of ATA as predictive of subsequent hypothyroidism (for example, Great Danes, Doberman Pinschers, and Old English Sheepdogs).

These thyroid panels and antibody tests can also be used for genetic screening of apparently healthy animals to evaluate their fitness for breeding. A bitch with antithyroid antibodies in her blood may pass these along to her puppies in her colostral milk. Also, any dog having circulating antithyroid antibodies can eventally develop clinical symptoms of thyroid or other autoimmune diseases. Therefore thyroid screening can be very important for potential breeding stock.

Thyroid testing for genetic screening purposes is less likely to be meaningful before puberty. Healthy young dogs (less than fifteen to eighteen months of age) should have thyroid baseline levels for all parameters in the upper third of the adult normal ranges. In fact, for optimum thyroid function in screening breeding stock, levels should be at least at the mid-point of the so-called "normal" ranges because lower levels may well be indicative of the early stages of thyroiditis among relatives of dog families known to have thyroid disease.

When treating with thyroid hormones: T4 is very forgiving, T3 can be easily toxic. With T4, dosage must be twice per day, as about half of it is metabolized and excreted from the body within twelve hours. For follow-up testing four to six weeks after initiating treatment, test at four to six hours after the morning dosage. For optimum levels, dogs should test in the upper third of the normal ranges after four to six weeks of therapy.

Dogs on long-term supplementation with thyroid hormones should be monitored with complete panels on a regular basis (every six to twelve months), and dosages should be adjusted accordingly.

Bibliography

1. Dodds, W.J. Hemostasis and blood coagulation. *Clinical Biochemistry of Domestic Animals*, 3rd edition, J.J. Kaneko (ed.), New York, Academic Press, 1980, pages 617-718, and 4th edition, 1989, pages 274-315.
2. Dodds, W.J.; Moynihan, A.C.; Fisher, T.M. and Trauner, D.B. The frequencies of inherited blood and eye diseases as determined by genetic screening programs. *JAAHA*, 17:697-704, 1981.
3. Dodds, W. J. von Willebrand's disease in dogs. *MVP*, 65:681-686, 1984.
4. Dodds, W. J. Genetic screening for hereditary bleeding disorders. *Kal-Kan Forum*, 1:52-58, 1982.
5. Dodds, W. J. Bleeding disorders. *Handbook of Small Animal Practice*. R. V. Morgan (ed.),Churchill Livingstone Inc., New York, NY, 1988, pages 773-786.
6. Dodds, W. J. Immune-mediated diseases of the blood. *Adv. Vet. Sci. Comp. Med.*, 27:163-196, 1983.
7. Dalton, D.G.; Savidge, G.F.; Matthews, K.B.; Dewar, M.S.; Kernoff, P.B.A.; Greaves, M. and Preston, F.E. Hypothyroidism as a cause of acquired von Willebrand's disease. *Lancet*, May 2: 1007-1009, 1987.
8. Ziegler, Z. R.; Hasiba, U.; Lewis, J.H.; Vagnacci, A.H.; West, V.A. and Bezek, E.A. Hemostatic defects in response to aspirin challenge in hypothyroidism. *Am. J. Hematol.*, 22:391-399, 1986.
9. Gosselin, S.J.; Capen, C.C.; Martin, S.L.; Krakowka, S. Autoimmune lymphocytic thyroiditis in dogs. *Vet. Immunol. Immunopathol*, 3:185-201, 1982.
10. Haines, D.M.; Lording, P.M.; Penhale, W.J. Survey of thyroglobulin autoantibodies in dogs. *AJVR*, 45:1493-1497, 1984.
11. Trence, D. L.; Morley, J. E.; Handwerger, B. S. Polyglandular autoimmune syndromes. *Am. J. Med.*, 77:107-116, 1984.
12. Fisher, D.A.; Pandian, M.R.; Carlton, E. Autoimmune thyroid disease: an expanding spectrum. *Pediatr. Clin. North Am.*, 34:907-918, 1987.
13. Larsson, M. Determination of free thyroxine and cholesterol as a new screening test for canine hypothyroidism. *JAAHA*, 24:209-217, 1988.
14. Dodds, W. J. Contributions and future directions of hemostasis research. *JAVMA*, 193:1157-1160, 1988.
15. Dodds, W. J. Bleeding and immune diseases, Parts I & II, and acquired von Willebrand's disease. Proc. 56th meeting AAHA, St. Louis,MO, 1989, pages 606-619.
16. Ciampolillo, A.; Mirakian, R.; Schulz, T.; Marini, V.; Buscema, M.; Pujol-Borrell, R. and Bottazzo, G. F. Retrovirus-like sequences in Graves' disease: implications for human autoimmunity. *Lancet*, May 20: 1096-1100, 1989.
17. Beale, K.M., Halliwell, R.E., and Chen, C.L. Prevalence of antithyroglobulin autoantibodies detected by enzyme-linked immunosorbent assay of canine serum. *JAVMA*, 196:745-748, 1990.
18. Bethune, J.E. Interpretation of thyroid function tests, *Disease-A-Month*, 35:541-595, 1989.

From: Bethune, John E. "Interpretation of thyroid function tests" Disease-A-Man Vol. 35, August 1989.

It is important to clearly understand the changes that occur in thyroid physiology during the evolution of clinical hypothyroidism from the euthyroid state in humans. As primary thyroid gland failure develops and progresses, the free T_4 concentration falls; in response to this, TSH is secreted from the pituitary gland, and its concentration in the serum rises. This stimulates the remaining thyroid tissue to make more T_4 so that its level does not fall below the lower limit of normal. This also prevents the signs and symptoms of hypothyroidism from developing. This state of "compensated" hypothyroidism may last for some time and can be detected only by the raised TSH value. As the thyroid gland is stimulated, the conversion rate of T_4 to T_3 within the gland increases, and sufficient extra T_3 is secreted to keep the level of free T_3 normal for a considerable time, even after the free T_4 level has fallen below the accepted normal range. If the patient is in a euthyroid sick state, however, the FT_3I may fall below normal. As the thyroid mass decreases with progression of the disease state, the T_4 concentration will finally fall below the normal range, and signs and symptoms of hypothyroidism will begin to appear. The T_3 level remains normal until very late in the development of hypothyroidism.

Autoimmune Diseases: The Failure of Self Tolerance

ANIMESH A. SINHA, M. THERESA LOPEZ, HUGH O. MCDEVITT

[Science, Vol. 248, June 15, 1990, pp. 1380-1388].

The Major Histocompatibility Complex in Autoimmunity

A large part of our understanding of the genetic basis of autoimmunity stems from the observation that susceptibility to many human diseases is associated with a particular MHC class I or class II allele. Of the more than 40 diseases known or thought to be autoimmune in nature, susceptibility to almost all is strongly influenced by genes encoded within the major histocompatibility complex (MHC), particularly the class I (HLA-A, B, C) and class II (Ia, HLA-D) MHC molecules.

Thus, even in individuals with a proven susceptible genotype, not all individuals will develop autoimmunity. The implication of this finding is that environmental factors also play a major role. The association of many autoimmune diseases with preceding infection raises the question of whether the initial autoimmune response might be triggered by an antigenically similar, cross-reacting environmental pathogen, a phenomenon often referred to as molecular mimicry.

A second important characteristic of autoimmunity is that susceptibility is polygenic. Because susceptibility is both multifactorial and polygenic, most autoimmune diseases are not inherited in a simple Mendelian segregation. The comparison of concordance rates for monozygotic twins with either HLA-identical dizygotic twins or HLA-identical siblings clearly indicates that MHC genes are an important but not a sufficient genetic factor in determining susceptibility to autoimmunity.

Environmental Agents Implicated in Autoimmunity

Numerous bacteria and viruses have been implicated in the etiology of autoimmune disease.

The mechanism of initiating autoimmunity may require that the infectious agent be present in the general population at a sufficient frequency to infect those individuals carrying the particular combination of susceptible HLA and non-HLA genes required for autoimmunity to develop.

In addition to the infectious agents discussed later, several drugs and toxins have been shown to precipitate autoimmune disease (for example, procainamide and SLE).

Perhaps the biggest challenge in the future will be the search for the environmental events that trigger self-reactivity.

Summary

In summary, autoimmunity (i) develops in genetically susceptible individuals, (ii) may be triggered by environmental agents operating by nonspecific inflammation or molecular mimicry, or both, (iii) is the result of the sum of genetic and environmental factors that overrides normal mechanisms of self tolerance, and (iv) is most often mediated by T cells or is characterized by an underlying defect or deregulation in the T cell compartment.

Autoimmune Thyroid Disease

W. Jean Dodds DVM

This article first appeared in Dog World, *April 1992.*

This article is intended as a followup to the excellent review of the thyroid gland by Dr. Edward Baker (November 1991 DW). Because so many breeds of dogs suffer from familial thyroid disease (Table 1), dog breeders and owners need to make a concerted effort to screen breeding stock for evidence of thyroid dysfunction before the dogs' use as stud dogs and brood bitches. This screening is important because their legacy carries the genetic material for the current and future decades. The information provided here outlines an approach that I have used successfully to reduce the prevalence of clinically expressed thyroid disease within susceptible families or breeds.

Most of the confusion about the diagnosis and treatment of thyroid disease in purebred or mixed-breed dogs today stems from the expectation that affected animals must show clinical signs of inadequate thyroid hormonal production (i.e., hypothyroidism) to have this disease. The term *hypothyroidism* has been loosely applied to describe all stages of this disease process, whereas strictly speaking it should be reserved for the end stages, when the animal's thyroid gland is no longer capable of producing sufficient hormone(s) to sustain clinical health. At this point, the dog can express any number of the nonspecific multisystem signs of thyroid dysfunction (Table 2). But let's start at the beginning.

The most common cause of canine thyroid disease is autoimmune thyroiditis (estimated 90 percent of cases). Thyroiditis is an immune-mediated process that develops in genetically susceptible individuals and is characterized by the presence of antithyroid antibodies in the blood or tissues. This condition usually, but not always, progresses to thyroid disease. (Other commonly used terms for the human disease are lymphocytic thyroiditis and Hashimoto's disease).

Thyroiditis is believed to start in most cases around puberty and gradually progresses through midlife and old age to become clinically expressed hypothyroidism once the thyroid glandular reserve has been depleted. During this process, the animal or person becomes more susceptible to immune-mediated or other diseases affecting various target tissues and organs—for example: autoimmune hemolytic anemia; idiopathic thrombocytopenic purpura; von Willebrand's disease; systemic lupus erythematosus; seizure disorders; chronic infections; such immunosuppressive viral infections as distemper, parvovirus, and retrovirus diseases; bone marrow failure; leukemia and lymphoma; chronic active hepatitis; immune eye, joint, kidney, and adrenal (Addison's) diseases; and chronic allergic and immune skin and muscle disorders. The prerequisite genetic basis for susceptibility to this disorder has been established in humans, dogs, and several other species.

To understand what happens in the transition from early thyroiditis to clinical hypothyroidism, a quotation from Dr. John E. Bethune, chairman of the UCLA Department of Medicine, is helpful (*Disease-A-Month*, 35, 1989).

"It is important to clearly understand the changes that occur in thyroid physiology during the evolution of clinical hypothyroidism from the euthyroid state in humans. As primary thyroid gland failure develops and progresses, the free T4 [l-thyroxine] concentration falls; in response, TSH [thyroid-stimulating hormone] is secreted from the pituitary gland, and its concentration in the serum rises. This rise stimulates the remaining thyroid tissue to make more T4 so that its level does not fall below the lower limit of normal. It also prevents the signs and symptoms of hypothyroidism from developing.

"This state of compensated hypothyroidism may last for some time and can be detected only by the raised TSH values. As the thyroid gland is stimulated, the conversion rate of T4 to T3 [triiodothyronine] within the gland increases, and sufficient extra T3 is secreted to keep the level of free T3 normal for a considerable time, even after the free T4 level has fallen below the accepted normal range. As the thyroid mass decreases with progression of the disease state, the T4 concentration will finally fall below the normal range, and signs and symptoms of hypothyroidism will begin to appear. The T3 level remains normal until very late in the development of hypothyroidism."

This explanation helps us to appreciate existing confusion and controversy within the veterinary profession about whether or not testing or treatment is indicated for dogs failing to show so-called typical signs of hypothyroidism.

In fact, we have only recently begun to recognize the subtle signs of early thyroid dysfunction in dogs as the prevalence of the autoimmune form of the condition has increased within and among dog breeds. Today, over 50 breeds are known to be genetically predisposed to develop thyroid disease (Table 1). Veterinary textbooks printed before 1990, and presumably even more recent ones, are not up to date on this topic; the latest scientific and medical discoveries are only now reaching refereed publications and have therefore not appeared in textbooks.

There are four main areas of examination when diagnosing thyroid disease.

• **Baseline thyroid profiles.** A typical thyroid test measures the total presence of the T4 hormone in the blood. But an important measurement is the small fraction of the total T4 that is not bound in the blood and is therefore available to the tissues. This "free" T4 is converted in the tissues to T3, which is important for intracellular function.

The newer thyroid profiles available at specialized veterinary laboratories in the United States and Canada test for total T4, total T3, free T4, cholesterol, and free T3. Cholesterol should be measured from the same blood sample drawn for the thyroid tests. In dogs with

Table 1

Breeds predisposed to thyroid and related immune-mediated diseases*

Afghan Hound	Irish Wolfhound
Airedale Terrier	Irish Setter
Akita	Keeshond
Alaskan Malamute	Kerry Blue Terrier
Australian Shepherd	Kuvasz
Basset Hound	Labrador Retriever
Beagle	Manchester Terrier
Belgian herding breeds	Mastiff
[Bernese Mountain Dog]	Miniature Schnauzer
Bouvier des Flandres	Newfoundland
Boxer	Old English Sheepdog
Briard	Pembroke Welsh Corgi
Bulldog	Pomeranian
Cairn Terrier	Poodle, especially Standard
Chow Chow	Rottweiler
Cocker Spaniel	Saluki
Collie, including Bearded	Samoyed
Dachshund	Scottish Terrier
Doberman Pinscher	Shar Pei
English Springer Spaniel	Shetland Sheepdog
English Setter	Shih Tzu
Fox Terrier	Siberian Husky
German Shepherd	Silky Terrier
Golden Retriever	Soft-Coated Wheaten Terrier
Gordon Setter	Vizsla
Great Dane	Weimaraner
Irish Water Spaniel	

**Probably an incomplete list.*

Table 2
Clinical Signs of Hypothyroidism*

alterations in cellular metabolism
lethargy
mental dullness
exercise intolerance
neurologic signs (polyneuropathy, seizures)
weight gain
cold intolerance
mood swings
hyperexcitability
stunted growth
chronic infections
hematologic disorders
bleeding
bone marrow failure
low red blood cell count (anemia)
low white blood cell count
low platelet count
ocular diseases
corneal lipid deposits
corneal ulceration
uveitis
keratoconjunctivitis sicca (dry eye)
infections of eyelid gland (meibomian gland)
neuromuscular problems
weakness
stiffness
laryngeal paralysis
facial paralysis
"tragic" expression
knuckling or dragging feet
muscle wasting
megaesophagus
head tilt
drooping eyelids
dermatologic diseases
dry, scaly skin and dandruff
coarse, dull coat
bilaterally symmetrical hair loss
"rat tail" or "puppy coat"
hyperpigmentation
seborrhea or greasy skin
pyoderma or skin infections
myxedema
chronic offensive skin odor
cardiac abnormalities
slow heart rate (bradycardia)
cardiac arrhythmias
cardiomyopathy
gastrointestinal disorders
constipation
diarrhea
vomiting
reproductive disorders
infertility of either sex
lack of libido
testicular atrophy
hypospermia
aspermia
prolonged interestrus interval
absence of heat cycles
silent heats
pseudo-pregnancy
weak, dying, or stillborn pups
other associated disorders
IgA deficiency
loss of smell (dysosmia)
loss of taste
glycosuria
chronic active hepatitis
adrenal endocrinopathies
pancreatic endocrinopathies
parathyroid endocrinopathies

**These signs appear during the end stages of thyroid disease.*

thyroid dysfunction, the higher the cholesterol level, the lower the level of free T4. Owners can obtain a printout listing all of the test results for their dogs, as well as the K value, which is a linear constant between free T4 and cholesterol.

Those K values more than +1 (and preferably more than +5) are considered to be normal. Less than -4 indicates an animal with primary thyroid disease, even if there are no overt symptoms. Values between -4 and +1 indicate either early thyroid disease or a nonthyroidal illness, which depresses thyroid levels. Thyroid deficiency even at early subclinical stages can cause irreversible damage. This more sensitive test is valuable because problems can be detected before damage has become so severe that symptoms are clear and visible.

For a dog displaying symptoms of autoimmune disease, the K value can be helpful to indicate whether the dog has low, normal, or high thyroid function, and whether the autoimmune problems are likely to be responsive to thyroid hormone supplementation. If test results are normal, and the dog is nevertheless ill, conduct other diagnostic tests, such as those for antithyroid antibodies (antithyroglobulin, anti-T4 and anti-T3 autoantibodies).

Michigan State University recently introduced antithyroid hormone antibody (anti-T4 and anti-T3) testing as part of its complete thyroid panel. This test is an important step toward more effective thyroid testing, and results of nearly 16,000 samples screened to date indicate a substantially higher prevalence than previously reported. (Contact the Endocrinology Section, Animal Health Diagnostic Laboratory, PO Box 30076, Lansing, MI 48909; 517-353-0621.)

• **Thyroid-stimulating hormone response test.** The thyroid profiling outlined above differs from the TSH response test often used to diagnose the *clinical* stages of hypothyroidism. This test measures thyroid reserve by assessing the ability of the thyroid gland to release T4 hormone on stimulation with an injection of TSH. It is estimated that from 50 percent to 70 percent of thyroid follicular tissue must be destroyed or damaged before hormonal reserves are depleted to the extent that overt hypothyroidism occurs. Thus, in the early stages of progressive thyroid disease (thyroiditis) the TSH response usually is normal and would be misleading—a false negative diagnosis. For this reason, we do not rely on finding a positive (abnormal) TSH response test to diagnose thyroiditis.

• **Antithyroglobulin antibody (ATA) test.** Thyroglobulin is a glycoprotein found in thyroid gland follicles. The metabolites of thyroid hormone, T3 and T4, are incorporated in the primary structure of thyroglobulin. Thyroglobulin is antigenic, and specific antibodies can form that are directed against several sites on the molecule. In humans, the presence of ATA is most often associated with Hashimoto's thyroiditis, the immune-mediated inflammation of the thyroid gland that progressively destroys functional thyroid tissue. The outcome of this process is hypothyroidism, and lifelong treatment with thyroid hormone is required. Hashimoto's thyroiditis is common; cases tend to cluster among families and certain ethnic groups. In humans, dogs, and other affected species, biopsy of the thyroid gland shows characteristic changes of lymphocytic infiltration and immunologic destruction.

Assays that measure ATA have been adapted for dogs. In two independent studies, about 50 percent to 80 percent of the clinically hypothyroid dogs tested had significant levels of ATA. Positive ATA titers were found also in some dogs with nonthyroidal endocrine disease, and in some euthyroid relatives of hypothyroid dogs. At present, the prevalence of ATA in the various breeds affected with hypothyroidism, the value of ATA as a predictor of impending thyroid failure or marker of familial hypothyroidism, and the relation of ATA to other immune-mediated disorders have not been determined. Preliminary studies of several affected dog families show promise in supporting the usefulness of ATA as predictive of subsequent hypothyroidism (e.g., Great Danes, Doberman Pinschers, Old English Sheepdogs, and Shetland Sheepdogs). The quantitative ATA test is offered at the veterinary college of the University of Florida in Gainesville, and as a research test for several groups, including Michigan State University.

• **Genetic screening for thyroid disease.** These thyroid panels and antibody tests can be used also for genetic screening of apparently healthy animals to evaluate their fitness for breeding. A bitch who has antithyroid antibodies in her blood may pass these along to her

puppies in her colostral milk. Also, any dog having circulating antithyroid antibodies can eventually develop clinical symptoms of thyroid or other autoimmune diseases. Therefore, thyroid screening can be very important for potential breeding stock.

Thyroid testing for genetic screening purposes is less likely to be meaningful before puberty. Screening is initiated, therefore, once healthy dogs and bitches have reached sexual maturity (between 10 and 14 months in males, and during the first anestrus period for females following their maiden heat).

Anestrus is a time when the female sexual cycle is quiescent, thereby removing any influence of sex hormones on baseline thyroid function. This period generally begins 12 weeks from the onset of the previous heat, and lasts one month or longer. The interpretation of results from baseline thyroid profiles in intact females is more reliable when they are tested in anestrus. Testing for health screening is performed at 12 to 16 weeks following the onset of the previous heat. In fact, genetic screening of intact females for other parameters, such as von Willebrand's disease, or wellness, health, and reproductive checkups also should be scheduled in anestrus females.

Once the initial thyroid profile is obtained, dogs and bitches should be rechecked annually to assess their health. Annual results permit comparisons that should reveal early evidence of developing thyroid disease or dysfunction. Annual testing also allows for early treatment where indicated to abort the development or advancement of clinical signs associated with hypothyroidism.

Healthy young dogs (younger than 15 to 18 months) should have thyroid baseline levels for all parameters in the upper half of the adult normal ranges. In fact, for optimum thyroid function in screening breeding stock, levels should be at least at the midpoint of the laboratory normal ranges. Lower levels may well be indicative of the early stages of thyroiditis among relatives of dog families known to have thyroid disease.

The new information summarized here has changed our approach to treatment and control of thyroid disease. We now know that in addition to providing thyroid supplementation for dogs showing the typical signs of thyroid disease, treatment of dogs showing the early stages of thyroiditis (based on the testing described above) is necessary to correct the underlying thyroid imbalance, reduce the risk of developing other related immune-mediated disorders, and prevent the process of thyroiditis from progressing to depletion and exhaustion of the thyroid gland.

The treatment of choice because of its wide safety margin and efficacy is T4 hormone (l-thyroxine or levothyroxine). The most commonly used brand names are Soloxine® (Daniels) and Synthroid (Flint). We recommend either of these products over generics, especially for the smaller breeds. T3 hormone (triiodothyronine) is not recommended for initial use because toxicity can more easily develop with this product. T3 is the intracellular hormone, whereas most of T4 must first be converted to T3 before it achieves its metabolic effect. In some cases, when the animal's body cannot properly convert T4 to T3, the dog will need both T4 and T3 therapy to correct the problem. For this purpose, the general rule of thumb is to give from two-thirds to a full dose of T4 and a one-third dosage of T3 (i.e., 0.1 mg of T4 per 10 to 20 pounds plus 1 ugm per pound of T3 twice daily). *However, no dog should be treated with these thyroid hormonal preparations without proper veterinary testing, medical examination, and followup.*

Thyroid hormones should always be given *twice* daily to effect the best response. Until recently, veterinarians were advised to treat either once or twice daily, because data on this point were unclear. We now know that the half-life of T4 in the dog is about 10 to 12 hours (much shorter than in humans); for T3, it's only 6 to 8 hours. Thus, about half of the hormone is metabolized and excreted from the body within 12 hours. Furthermore, twice-daily dosing aids in controlling thyroiditis because it shuts off pituitary production of TSH by negative feedback in concert with the half-life of the hormone. In other words, the dog's own thyroid follicular cells become quiescent and are less likely to stimulate production of the antithyroid antibodies responsible for the disease. (Obviously, these are simplistic explanations of the complex metabolic, immunologic, and biochemical events involved.)

Contrary to some popular wisdom, treatment with thyroid hormone does not destroy or suppress the potential of the gland to respond on its own once treatment is stopped for whatever reason. The latest veterinary research shows that the thyroid gland requires at least 30 days to recover its full hormonal potential once therapy is withdrawn. Therefore, if an animal has been medicated and the diagnosis is unclear, treatment should be withdrawn (if it's clinically safe to do so) for 30 days before the animal is retested with the complete type of thyroid profile described above.

Followup testing after initiating treatment usually is performed after four to eight weeks of therapy. The sample should be taken four to six hours after the morning dosage; optimum results will show thyroid values in the upper third of normal ranges at that peak time of absorption. Dosage can then be adjusted accordingly if needed. Dogs on long-term therapy with thyroid hormones should be monitored with complete panels (not just T4, because you need to be sure that the dog's body is properly converting the T4 medication to T3) every 6 to 12 months.

Additional reading

Dodds, W. J. "1990 Update on Autoimmune Disorders of Purebred Dogs." *Pure-Bred Dogs/American Kennel Gazette,* Vol. 107, No. 7 (July), 86-94, 1990. [See also *The Alpenhorn*, February 1991, p. 29.]

The author is president of HEMOPET, Santa Monica, California, and director of the Comparative Hematology Laboratory, New York State Department of Health.

Thyroid Can Alter Behavior

W. Jean Dodds DVM

This article first appeared in Dog World, *October 1992.*

Abnormal behavior in dogs can have a variety of medical causes; it also can reflect underlying problems of a psychological nature. Your veterinarian follows a systematic diagnostic approach in searching for medical causes when a pet exhibits unusual or unacceptable behavior. As summarized by Landsberg (*Canadian Veterinary Journal*, 31:225-227, 1990), this approach includes:

a complete patient history;

2. clinical examination and a neurological workup;
3. routine laboratory testing of complete blood count, blood biochemistry and thyroid profiles, urinalysis, fecal exam, and X ray;
4. additional specific laboratory tests as indicated-(e.g., other hormonal tests, bile acids, blood ammonia, glucose tolerance, immunologic assays, and tests for toxins, fungi, and other infections);
5. examination of cerebral spinal fluid; and
6. more specialized neurological examinations, such as an electroencephalography and computerized axial tomography scan.

Diagnostic steps 1 through 3 are usually completed first; additional tests, such as steps 4 through 6, are performed if indicated. If these test results prove to be negative, a veterinary behavior consultant or qualified pet behavior therapist should evaluate the dog.

Inheritance has been shown to play an important part in the behavior of both animals and humans; Plomin recently reviewed its role (*Science*, 248:183-188, 1990). The genetic influence on behav-

ioral disorders rarely accounts for more than half of the phenotypic expression of behavioral differences. Each of the multiple genes involved has a small effect on behavior. Newer techniques in molecular biology should permit the identification of the genetic DNA marker sequences responsible for behavioral variation.

However, behavior is the most complex phenotype because it not only reflects the functioning of the whole organism but also is dynamic and changes in response to environmental influences. In regard to animal behavior, applied behavioral genetics first was studied several thousand years ago, because animals were bred and selected as much for their behavior as for their conformation. The dramatic differences in behavior and physique among various dog breeds attest to the results. Today, these breeds have a great range of genetic and behavioral variability.

In recent years, many investigators have noted the sudden onset of behavioral changes in dogs around the time of puberty. Most of the dogs have been purebreds or crossbreds with an apparent predilection for certain breeds (e.g., Golden Retrievers, Shetland Sheepdogs, German Shepherd Dogs, Cocker Spaniels, Akitas, Doberman Pinschers, Rottweilers). Many of these dogs also had begun to show the seasonal effects of allergies to inhalants and ectoparasites, such as fleas, followed by the onset of skin and coat disorders, including pyoderma, allergic dermatitis, alopecia, and intense itching.

A typical history starts with a quiet, well-mannered, sweet-natured puppy. The dog is outgoing; attended training classes to prepare for obedience, working, or show events; and had a reputable breeder whose kennel has no history of behavioral problems.

However, at the onset of puberty, which varies from 7 months to 1 year, sudden major personality changes are observed. Typical signs may include incessant whining, nervousness, schizoid behavior, fear in the presence of strangers, hyperventilation, undue sweating, occasional disorientation, and failure to be attentive. These symptoms can progress to sudden unprovoked aggressiveness in unfamiliar situations with other animals and with people, especially children.

The owners may attribute the problems to the sex hormonal changes accompanying puberty or just the uncertainties of adolescent development. Often, these animals are neutered, which appears to alleviate the behavioral problems, specifically the aggression, for varying lengths of time. In a significant proportion of these animals, however, neutering does not alter the symptoms, and they intensify progressively to the point that the adult can be described as flaky, unable to handle any kind of stress, frantically circling, hyperventilating, and not able to settle down. Animals used for field work and tracking often fail to follow the scent, whereas those in obedience training may lose the scent articles. Their powers of concentration are often very short, so dogs that were training very successfully in obedience appear to lag behind in a disinterested fashion. With all of these changes in behavior, the problem of most concern is unwarranted aggression. When large breeds are affected, it poses a significant hazard to family members, friends, and strangers.

In some cases, affected animals do not show aggression but become very shy and fearful to the point that they are social outcasts and do not make acceptable house pets. These animals clearly are not suitable for show, obedience, or working purposes. Some of these dogs will show extremely submissive behavior, roll over, and urinate on being approached.

The third group of dogs showing aberrant behavior consists of those that experience seizure or seizure-like disorders beginning in puberty and continuing to midlife. These are dogs that appear perfectly healthy outwardly and have normal hair coats and energy, but suddenly experience seizures for no apparent reason. The seizures are often spaced several weeks to months apart, and occasionally they appear in a brief epileptiform cluster. In some cases, the animals become aggressive and attack those around them shortly before or after having one of these seizure episodes.

The number of dogs showing various types of abnormal behavior in these three classical modes (aggression, extreme shyness, or seizure-like activity) has been increasing over the last decade. Veterinary colleagues have remarked that in recent years some young dogs have become completely unacceptable because of bizarre, sudden behavioral changes. Consequently, we began to examine these animals by using the stepwise diagnostic approach outlined. The importance of performing complete laboratory profiles on the blood and urine, with specific emphasis on thyroid hormonal function, was stressed.

We were surprised to find that in many of the cases studied, significant abnormalities were found in the thyroid profile. Some cases also had changes in the liver enzyme patterns, specifically with abnormal increases in pre- and postmeal bile acids and elevated gamma glutamyl transferase levels. About 10 percent of these young dogs had abnormalities of the liver profile, and a few also had changes in renal function. For the majority, however, the primary abnormality was attributable to abnormal thyroid function. This thyroid dysfunction would classically express high levels of T3 (triiodothyronine) and T4 (l-thyroxine) autoantibodies, with an artifactual, apparent elevation of total T3 level. It would not be uncommon to find circulating total T3 levels that read as much as 3,000 to 5,000 nanograms per deciliter. Although not all of the affected animals had documented evidence of T3 and T4 autoantibodies, some had positive antithyroglobulin antibody tests. In either event, the diagnosis was confirmed as autoimmune thyroiditis.

The autoimmune thyroid disease present in these patients apparently is inducing some type of physiological change at the cellular level, which leads to their aberrant behavior. This supposition can be made with some assurance because treatment of thyroiditis in these dogs with appropriate doses of thyroid replacement hormone twice daily, along with a one-month tapering course of low-dose corticosteroids, has successfully reversed the behavioral problems within four to eight weeks. Dramatic changes in behavior have been recognized in a few cases as early as after 10 days of therapy.

By contrast, it usually takes five to seven months of thyroid replacement therapy to effect complete disappearance of the circulating antithyroid antibodies. These dogs should be maintained for life on the appropriate dose of thyroid hormone, which may need to be adjusted periodically.

Another subset of affected dogs consists of those that do not have demonstrable antithyroid antibodies but have baseline thyroid profiles that are clearly abnormal. In these cases, levels of total T4, total T3, free T4, and free T3 are usually well below the lowest limits of the adult normal ranges, or are in the low normal or borderline ranges. The second situation is of particular significance in young dogs 9 to 15 months old. When these dogs are treated with standard doses of thyroid replacement therapy (0.1 milliliter per 10 pounds of body weight twice daily), the clinical signs associated with abnormal behavior rapidly resolve.

To date, more than 25 animals have been diagnosed as having thyroid imbalance as the major, if not exclusive, cause of their behavioral abnormalities. Somes have been followed for as long as three years and are still exhibiting normal behavior. Animals on therapy returned successfully to obedience activities, completed show championships, and undertaken active field and tracking work.

Here are two case studies as examples.

In the first, a 4-year-old male Akita, weighing 110 pounds, suddenly attacked his owner and bit her in the face. The dog had been in the same family since early puppyhood, and had been a remarkably even-tempered, well-behaved, and nonaggressive pet with people and other animals. After seeking the advice of two different veterinary clinicians and a major teaching hospital, the owner was about to give up in despair because no physical abnormalities could be found.

She was referred to us by an Akita rescue group, as this pattern of

behavioral change has been associated with thyroid dysfunction in the breed. A complete thyroid panel, which had not been performed earlier, was suggested, and the dog was found to be hypothyroid. Thyroid therapy was initiated on a twice-daily basis. The dog's exemplary temperament returned, and he has not shown any unusual behavior for more than a year. An interesting additional complication of the case was a moderately severe thrombocytopenia, which also resolved with low doses (5 to 10 milligrams) of alternate-day steroid therapy and thyroid medication.

In the second case, a 9-month-old male Shetland Sheepdog from excellent show-quality bloodlines suddenly became frantic and fearful. Acting intermittently, as if his vision were impaired, he attacked a toddler in the owners' home. A complete physical examination and laboratory testing were conducted; a routine check for T4 was borderline normal. The dog's abnormal behavior appeared to resolve but soon reappeared.

After a second attack, a complete thyroid profile was performed at Michigan State University's Animal Health Diagnostic Laboratory (telephone 517-353-0621). The total T4 was 44 nmol/l, total T3 was 0 nmol/l, free T4 was 2 pmol/l, free T3 was >20 pmol/l, T4 autoantibody was 18, and T3 autoantibody was 85. The referring veterinarian did not realize that the results were consistent with autoimmune thyroiditis, and the dog was not treated. Two months later, the dog attacked another person, and a second thyroid profile was sent to the Michigan State laboratory. The second profile showed a total T4 of 29 nmol/l, total T3 of 0 nmol/l, free T4 of 25 pmol/l, free T3 of >20 pmol/l, T4 autoantibody of 48, and T3 autoantibody of 91. At this point, our group was consulted.

The dog had a previous history of facial demodectic mange. Because corticosteroids are not recommended with demodecosis, the treatment consisted of a full therapeutic dose of T4 thyroid supplement at 0.1 milligram per 10 pounds and a one-third dose of T3 thyroid supplement at 1 microgram per pound, both twice daily. The rationale for treating with both T4 and T3 supplements in this case was to attempt to normalize the thyroid axis as quickly as possible to avoid danger to family members. At the time of this writing, the dog's behavioral aggression has subsided.

For those animals that show occasional seizure disorders, thyroid medication alone usually will suffice. Anticonvulsant medication is needed along with the thyroid therapy to control cases with more severe seizure clusters. The anticonvulsant of choice would be phenobarbital or, alternatively, sodium bromide, particularly if the patient has abnormalities of liver function.

Because many of these animals have autoimmune thyroid disease, concomitant medical management includes avoiding environmental factors that can further challenge the immune system. That means placing the animal on a hypoallergenic "natural" diet preserved with vitamins E and C (e.g., lamb-and-rice-based, lower-protein kibble without added chemical preservatives); avoiding drugs, such as the potentiated sulfonamide antibiotics and monthly heartworm preventives that may adversely affect the immune system in these susceptible dogs; and withholding vaccination boosters until the thyroid function is balanced properly and the behavioral abnormalities are resolved. If animals are due for annual vaccine boosters during this period, vaccine antibody titers for distemper and parvovirus can be determined.

If your otherwise healthy young or adult dog experiences sudden behavioral changes, you should consult your vet and check for an underlying thyroid imbalance as shown by:

1. the presence of thyroid autoantibodies;
2. low or borderline levels of total T4, total T3, and free T4; or
3. failure to triple baseline total T4 levels in response to challenge with thyroid-stimulating hormone. In our experience, the most predictive thyroid test parameters to identify these cases are 1 and 2, because the TSH response test just measures thyroid reserve, which remains adequate in the early stages of thyroid disease.

An earlier version of this article appeared in DVM Newsmagazine, Vol. *23, No. 5, 1992. The author is president of HEMOPET in Santa Monica, California, and director of the Comparative Hematology Laboratory, New York State Department of Health.*

W. Jean Dodds DVM

a Seminar

on CANINE THYROID and AUTOIMMUNE DISORDERS

Sunday, June 23, 1996
The Embassy Suites Hotel
Markham, ON Canada

Dr. Jean Dodds

PROFESSIONAL BIOGRAPHY

Dr. Dodds was born in Shanghai, China on January 4, 1941. She received her D.V.M. in 1964 from the Ontario Veterinary College. In 1965 she moved to the New York State Health Department in Albany and began comparative studies of animals with inherited and acquired bleeding diseases. This work continued full-time until 1986 when she moved to Southern California to establish Hemopet, the first nonprofit national blood bank program for animals.

From 1965-1986, shewas a member of many national and international committees on hematology, animal models of human disease, veterinary medicine, and laboratory animal science. Dr. Dodds was a grantee of the National Heart, Lung, and Blood Institute (NIH) and has over 150 research publications. She was formerly President of the Scientist's Center for Animal Welfare, and Chairman of the Committee on Veterinary Medical Sciences and Vice-Chairman of the Institute of Laboratory Animal Resources, National Academy of Sciences. In 1974 Dr. Dodds was selected as Outstanding Woman Veterinarian of the Year, AVMA Annual Meeting, Denver, Colorado; in 1977 received the Region I Award for Outstanding Service to the Veterinary Profession from the American Animal Hospital Association, Cherry Hill, New Jersey; in 1978 and 1990 received the Gaines Fido Award as Dogdom's Woman of the Year; and the Award of Merit in 1978 in Recognition of Special Contributions to the Veterinary Profession from the American Animal Hospital Association, Salt Lake City, Utah. In 1984 she was awarded the Centennial Medal from the University of Pennsylvania School of Veterinary Medicine. In 1987 she was elected a distinguished Practitioner of the National Academy of Practice in Veterinary Medicine. In 1994 she was given the Holistic Veterinarian of the Year Award from the American Holistic Veterinary Medical Association, Orlando, Florida. She is a member of numerous professional societies.

Today, Dr. Dodds is actively expanding Hemopet's range of nonprofit services and educational activities. The animal blood bank program provides canine blood components, blood bank supplies, and related services throughout North America. Hemopet's retired Greyhound blood donors are adopted as pets through the Pet Life-Line arm of the project. On behalf of Hemopet, she consults in clinical pathology nationally and internationally, and regularly travels to teach animal health care professionals, companion animal fanciers, and pet owners on hematology and blood banking, immunology, endocrinology, nutrition and holistic medicine. She is also the Editor of *Advances in Veterinary Science and Comparative Medicine* for Academic Press.

Why Should I .... ?

"Why should I as a purebred dog owner, or breeder, or veterinarian or vet-tech, be attending the W.Jean Dodds Seminar this June?"

Here's just a sampling of why ....

DIAGNOSIS AND MANAGEMENT OF CANINE THYROID DISEASE

W. Jean Dodds, DVM

Although thyroid dysfunction is the most frequently recognized endocrine disorder of the dog, it is sometimes difficult to make a definitive diagnosis. Many clinical signs of thyroid dysfunction mimic symptoms resulting from other causes so that recognition of the problem and interpretation of results of thyroid function tests can be problematical. The majority of canine thyroid disease is due to autoimmune thyroiditis, which is a familial disorder of inherited predisposition similar to that of human Hashimoto's disease. Therefore, the most complete approach to thyroid testing should include assays for thyroid autoantibodies.

1. Baseline Thyroid Profiles

Because of the difficulties inherent to diagnosing thyroid disease, the use of a complete baseline thyroid panel is advisable and should include measurements of total T4, total T3, free T4, free T3, and circulating T4 and T3 autoantibodies. This type of profile can be applied not only to clinical patients suspected of having thyroid disease, but also can be used for genetic screening of apparently healthy relatives to evaluate their fitness for breeding. A bitch with circulating antithyroid antibodies can pass these along to the puppies transplacentally as well as via the colostrum. Furthermore, any dog having circulating antithyroid autoantibodies may eventually develop clinical symptoms of thyroid disease and/or be susceptible to other autoimmune diseases. Thyroid prescreening is thus very important for selecting potential breeding stock.

2. Genetic Screening for Thyroid Disease

Thyroid testing for genetic screening purposes is less likely to be meaningful before puberty. Screening is initiated, therefore, once healthy dogs and bitches have reached sexual maturity (between 10 to 14 months in males and during the first anestrous period for females following their maiden heat). As the female sexual cycle is quiescent during anestrus, any influence of sex hormones on baseline thyroid function will be avoided. This period generally begins 12 weeks from the onset of the previous heat and lasts one month or longer. The interpretation of results from baseline thyroid profiles in intact females will be more reliable when they are tested in anestrus. In fact, genetic screening of intact females for other disorders such as von Willebrand's disease, hip dysplasia, inherited eye diseases, and wellness or reproductive checkups is best scheduled during anestrus. Once the initial thyroid profile is obtained, dogs and bitches should be rechecked on an annual basis to assess their thyroid function and overall health. Generation of annual test results provides comparisons that permit early recognition of developing thyroid dysfunction. This allows for early treatment, where indicated, to avoid the appearance of advancement of clinical signs associated with hypothyroidism.

For optimal health, young dogs under 15 to 18 months of age should have thyroid baseline levels in the middle to upper half of adult normal ranges. This is because puppies and adolescent dogs are still growing and maturing, and require higher levels of thyroid hormones to promote development. Similarly, in older animals above 8 or 9 years of age, body functions slow down and so baseline thyroid levels may be in the lower third of the range in the euthyroid individual. For healthy young adults used for performance or breeding, optimum thyroid function should be at least at the mid-point of the laboratory normal ranges. Lower levels may be indicative of the early stages of thyroiditis among relatives of dog families previously documented to have thyroid disease.

3. Diagnosing Difficult or Equivocal Cases

The difficulty in accurately diagnosing some cases of thyroid disease is compounded by the fact that some patients with typical clinical signs of hypothyroidism have circulating levels of thyroid hormones within the normal range. Most of these patients will improve clinically when given thyroid medication. This is because the circulating blood levels of thyroid hormones may not reflect the cellular and tissue levels of these same hormones. A 6 to 8 week clinical trial of thyroid supplementation given twice daily is safe and appropriate for such patients, and is followed by rechecking the complete thyroid profile 4 to 6 hours after the morning pill. Response to thyroid therapy is considered an appropriate justification for continuing to prescribe thyroid hormone along with annual checkups to adjust dosages as appropriate.

In the event that an animal is already receiving thyroid supplementation and the basis for the original diagnosis is unclear, the clinician may elect to discontinue therapy and retest. In such cases, or when thyroid therapy is discontinued for any other reason, retesting is performed after another 4 or preferably 6 weeks. This is because it takes at least a month for the animal's own pituitary-thyroid axis to be restored to full productive capacity after cessation of thyroid therapy. To perform thyroid retesting before this period has elapsed would yield relatively low blood levels of thyroid hormones which would bias interpretation of test results in favor of hypothyroidism.

Once the appropriate dose of thyroid supplement is established, annual retesting is recommended unless the dog exhibits any sign of illness in the intervening period.

DIAGNOSTIC TESTING FOR THYROID DISEASE

W. Jean Dodds, DVM
HEMOPET
938 Stanford Street
Santa Monica, CA 90403
(310) 828-4804; FAX (310) 828-8251

Canine hypothyroidism and feline hyperthyroidism are the most common endocrine disorders of companion animals. A recent survey of breed clubs conducted by the American Kennel Club Delegates Committee on health matters indicated that hypothyroidism was the most common health problem of concern to the majority of purebred dog fanciers. However, confusion remains over which diagnostic tests are the most specific and sensitive for identifying thyroid dysfunction especially in its early stages. Accurate diagnosis of the early compensatory stages of canine hypothyroidism and autoimmune thyroiditis, as well as feline hyperthyroidism, affords important clinical consequences for early therapeutic intervention and case management. Additionally, the heritable nature of autoimmune thyroiditis in dogs has significant genetic implications for breeding stock.

Older Tests for Thyroid Disease

Measuring serum T4 (or T3) alone is not a reliable means for diagnosis of thyroid disease in any species, because it can : 1) overdiagnose hypothyroidism; 2) underdiagnose hyperthyroidism; and 3) may fail to detect early stages of the compensatory disease and thyroiditis. These tests are greatly influenced (lowered) by the presence of nonthyroidal illness (NTI) and specific drug therapy (e.g. corticosteroids, anticonvulsants, nonsteroidal anti- inflammatory agents). For these reasons, measuring serum T4 alone or as the only thyroid analyte of a health profile is not recommended.

Current and New Tests for Thyroid Disease

Free (Unbound) T4

Clinical veterinary diagnostic laboratories such as the MSU Animal Health Diagnostic Laboratory, Professional Animal Laboratory, and Veterinary Research Laboratories in the United States, and VitaTech in Canada, have continued to offer state-of-the-art diagnostic tests for thyroid disease. The recommended panels include total T4, total T3, free T4, free T3, T3 autoantibody and T4 autoantibody. Accurate measurement of free T4 is an important component of this analysis because it represents the biologically active (unbound) fraction

of the total T4. Methodologies for measuring free T4 have used both a solid-phase analog assay and equilibrium dialysis (EQD), the latter being considered the "gold standard" against which the solid-phase analog assay has been validated. In keeping abreast with the emerging technologies, chemiluminescence solid-phase methods for measuring free T4 have been developed which are easier to perform than radioimmunoassays, and have a very high degree of correlation (>96%) with EQD.

Free T4 assays are less likely to be influenced by NTI and drug therapy. Reliable (validated) methods for measuring this analyte are very useful for proper diagnosis of canine hypothyroidism and feline hyperthyroidism, especially in the early stages or in the presence of NTI.

Endogenous Canine Thyroid Stimulating Hormone (cTSH)

Validated assays for cTSH are now available. In primary hypothyroidism, as serum free T4 levels fall, pituitary output of TSH rises in a regulatory, compensatory response. Elevated serum TSH levels are therefore an important indicator of thyroid dysfunction. When measured as part of a complete thyroid panel or along with free T4, the cTSH assay offers another analyte to more precisely assess canine thyroid function. Recent experience with these tests, however, indicates about 10-15% discordancy between expected and actual results in normal dogs as well as confirmed cases of hypothyroidism or NTI. In humans with chronic renal disease (CRD), for example, there is a much higher incidence of true hypothyroidism than in age-matched control people, but in other euthyroid CRD patients TSH levels are also increased (Kaptein, EM. End. Rev. 17: 45-63, 1996).

Canine Thyroglobulin Autoantibodies (TgAA)

An estimated 80% of cases of canine hypothyroidism result from heritable autoimmune (lymphocytic) thyroiditis. In most of these cases, TgAA are present in the serum, whereas only about 20% of cases of thyroiditis have elevated circulating T3 and/or T4 autoantibodies. Thus, the presence of elevated T3 and/or T4 autoantibodies confirms a diagnosis of autoimmune thyroiditis but underestimates its prevalence, as negative (non-elevated) autoantibody levels do not rule out thyroiditis.

The presence of elevated TgAA levels confirms thyroiditis, promotes early recognition of the disorder, and facilitates genetic counselling. A commercial test for measuring TgAA has just been introduced and awaits validation with a representative number of field cases from the various breeds affected with autoimmune thyroid disease.

References

[specific references on topics about thyroid disease are available upon request. The literature base is extensive and changing rapidly]

ENDOGENOUS CANINE TSH

W. Jean Dodds, DVM
HEMOPET
938 Stanford Street
Santa Monica, CA 90403
(310) 828-4804 Fax (310) 828-8251

Reliable testing for endogenous canine thyroid stimulating hormone (cTSH) is now commercially available. This important new assay to assess canine thyroid function is best used in conjunction with a regular baseline canine thyroid antibody profile (measuring total T4, total T3, free T4, free T3, and T4 and T3 circulating autoantibodies).

The anterior pituitary gland produces TSH, which, in turn, acts upon the thyroid gland to regulate circulating levels of T4 and T3 hormones. Production and secretion of TSH is controlled by negative feedback from circulating T4 and T3 levels, as well as by thyroid releasing hormone (TRH) produced by the hypothalamus. When circulating levels of thyroid hormones go down or are inadequate to maintain normal metabolic demands, TSH output increases and circulating levels of TSH rise. The higher the elevation of endogenous TSH beyond basal levels, the lower is the test animal's thyroid glandular output.

Primary hypothyroidism impairs production of thyroid hormones and so elevated levels of endogenous TSH are found. By contrast, in secondary or tertiary hypothyroidism, where thyroid hormone production is low because of hypothalamic or pituitary disease or is suppresses by nonthyroidal illnesses, lowered or normal levels of endogenous TSH are expected.

While the cTSH level alone or in conjunction with a validated freeT4 level can be used to assess canine thyroid function, we do not recommend this approach for routine diagnostic workup because dogs having circulating T3 and /or T4 autoantibodies (as an indicator of autoimmune thyroiditis) would not be identified. Knowing that a canine patient or healthy relative exhibits circulating thyroid autoantibodies has important implications for management and treatment followup, especially if only T3 and T4 are monitored, as well as for genetic counselling about the advisability of removing these animals from a breeding program.

Including the cTSH assay in thyroid profiling can more definitively identify cases of primary hypothyroidism. It can also be used to monitor the response of hypothyroid dogs to thyroid therapy, because the elevated levels initially seen should come down into the normal reference range as the levels of thyroid hormones become normalized and clinical symptoms wave. [Williams, DA et al, J Vet Int Med 9(3): 184, 1995 (abstr.); Nachreiner, RF et al, ibid, 1995 (abstr.)].

DVM Newsmagazine Vol. 22 #4,
April 1991 pp. 15,18

Enhancing Immune Competence with Nutritional And Other Approaches

by
W. Jean Dodds, DVM

A normal functioning immune system is an essential component to promote the health and longevity of humans and animals. The immune system is a delicately balanced and integrated functional network of humoral and cellular components that protect the organism against acute and chronic disease. Stress is a major influence on this function and can be physiological, physical and emotional. Immune function tends to be more active in the young, stable in mid-life and then wanes gradually in old age. Throughout life, immunological recognition and reactivity is continually challenged by an array of environmental agents that serve to enhance failure of self-tolerance which produces autoimmunity by mechanisms of non-specific inflammation and/or molecular mimicry. Molecular mimicry is the process whereby various viruses, bacteria, chemicals, drugs and other toxins challenge the immune system and produce antibodies that at the same time recognize similar antigenic components in self tissues. These antibodies are then capable of reacting and inhibiting or destroying these like tissues. As described in the last article in this series (December 1990 issue of DVM Magazine) the environmental effects on the immune system are only realized in genetically susceptible individuals. The events that trigger self-reactivity or auto-immunity result from the sum of both the genetic and the environmental factors involved. These reactions are most often mediated by T-lymphocytes or by their dysfunction.

The genetic influence on immune dysfunction is perhaps best exemplified by purebred animals where line breeding and inbreeding facilitates both the transmission and the recognition of genetic defects once the mutation has arisen. Genetic mutations themselves are more likely to occur when inbreeding is practiced. As discussed in earlier articles in this series, large scale screening programs for the identification of affected and/or carrier animals within the population at large are an effective mechanism to identify and eventually control the frequency of these defects. Depending on the mode of inheritance, different methodologies need to be applied for the detection and control of genetic diseases. However, coupled with this approach to eliminating undesirable traits is the necessity to monitor breeding stock regularly for overall soundness, reproductive health and performance.

Application of molecular biology and genetic engineering principles to the identification of the genes involved in transmitting common diseases to purebred animals is an exciting new approach. The laboratory techniques to perform these genetic studies have been developed. Their application to map animal genomes, while complex and time consuming, is

feasible. Over the next five to ten years, important information will no doubt evolve about the specific genes that cause some of the more common canine diseases. In the meantime, it is important that the top producing sires and foundation bitches of a particular disease be screened for these diseases because they represent the major nucleus of genetic material for the current and future decades. Once these probes are available, however, it remains to be seen whether application of these techniques will in fact provide a practical and relatively simple and affordable means of screening breeding stock. Despite the proven benefits of this approach and intensive educational efforts to encourage its use, it must be recognized that some breeders will choose not to participate. Veterinarians and scientists who provide testing and counseling services of this nature need to be understanding in dealing with the concerns and sensitivities of these individuals. Success with these programs depends upon an honest, voluntary commitment to the principles and ethics of breeding better companion animals.

For the most common genetic defects of pure bred dogs, there is another potential stumbling block to the effective implementation of these molecular probes in the elimination of genetic defects. If the majority of the best foundation stock of a breed is carrying the gene for an undesirable trait, then removing those animals from the breeding pool will result in the necessity of inbreeding and line breeding the remaining animals who may be less desirable for a variety of other reasons. Obviously, whether or not this concern impacts a breed in question will depend upon the nature of the disease, its clinical severity and its prevalence. Whenever this type of situation exists, an alternative approach would be to accept that there is a widespread influence of a particular gene within a breed and to concentrate instead on reducing the environmental influences that trigger the genetic susceptibility to a particular disorder. This approach would only work for the complex diseases where several genes are involved in controlling disease expression. It is recognized today that most complex diseases of humans depend upon four or perhaps five genes interacting together. It is the environmental influences on these genes that cause the genetic susceptibility to be expressed clinically.

Environmental Influences on Immune Function

One obvious way to reduce the environmental effects on genetic susceptibility to disease is to reduce or eliminate environmental factors. Examples here would be reducing environmental exposure to toxins such as pesticides, both in the physical environment and applied to animals topically, reduce the intake of drugs used on a regular basis for prevention of disease, whenever feasible alternatives apply and basically accepting that as a society, we need to adopt a more ecologically sound approach to conservation of the environment and in reducing pollution. These principles apply not only to companion and

domestic animals, but also to ourselves as we share a common environment. In fact, companion animals provide an ideal sentinel for the effects of the environment on disease, especially because their genetic makeup is more alike.

If we are able to reduce the environmental factors that promote disease in animals, then by doing so we should enhance competence of the immune system and its ability to fight off or cope with environmental challenges. One of the most important ways to reduce continual immunological challenge which could suppress overall immune function is through the diet. Food stuffs ingested by animals on a regular basis may be either imbalanced in terms of major nutrients, minerals and vitamins and often contain chemicals that are added to the final product to enhance its stability and shelf life. The question we need to address is whether the content and/or the addition of chemical preservatives to animal foods is potentially dangerous to those animals genetically predisposed and already susceptible to the immunological challenge which can promote autoimmune disease. An example is the current controversy concerning the preservative ethoxyquin. This preservative has been blamed for most of the chronic maladies that have been affecting purebred dogs with increasing frequency. The nearly ten fold increase in the frequency and recognition of immune-mediated diseases in companion animals has coincided with the introduction of ethoxyquin into the premium brands of pet foods. Whether or not there is any cause and effect relationship here remains to be determined. However, to date there are no documented studies that assure the safety of ethoxyquin chronically fed to dogs and cats at the levels currently approved for addition to animal food by the Food and Drug Administration. To be fair, there are also no data that show ethoxyquin to be unsafe when fed at the currently approved levels. A new chronic feeding trial in dogs is currently underway by the Monsanto Company that manufactures this preservative. We can hope that data generated from this new scientifically controlled study will shed some light on the question. In the meantime, veterinarians and purebred animal breeders and owners are faced with a dilemma of how to deal with nutritional issues that apply to a genetically susceptible subpopulation of purebred animals.

Many of us that have been dealing with management and treatment of purebred animals suffering from immune-mediated diseases have taken the approach that eliminating environmental challenges includes the necessity of alternative nutritional management. The results have been remarkable. Nutritional support has been used as an adjunct and essential component to the treatment of these diseases along with optimizing laboratory testing, monitoring, and treating where appropriate with immunosuppressive drugs; hormonal supplementation for the autoimmune endocrine and reproductive disorders; bone marrow stimulants and antibiotics. The animals are placed on an elimination diet for three or more weeks as would be done when a food allergy or hypersensitivity is suspected. Standard

commercial diets containing chemical preservatives are replaced with the so-called "natural" or hypoallergenic diets that do not contain added chemical preservatives beyond what is present in trace amounts in the raw ingredients. Alternatively, if feasible, the animal is placed on a homemade diet using one of the formulas readily available to treat animals depending on the specific disease involved. All other food supplements including treats are removed, with the exception of fresh or stewed vegetables, low-fat cottage cheese or plain non-fat yogurt as desired by the owner to enhance palatability. A mixture of fresh vegetables, excluding onions and the cabbages, is stewed together with garlic and fresh or dried mixed Italian herbs. This is used as a vegetable soup mixture to add to the basic kibbled cereal. The cottage cheese and yogurt dairy products are avoided for those animals having a specific intolerance to these foods. Other additives can include boiled eggs, stewed chicken or lamb with the fat skimmed off, steamed brown rice and pasta. [Obviously these are just examples of the approach used and individual diets can be developed by owners in consultation with their veterinarian and a nutritionist].

In our experience, this nutritional approach in conjunction with conventional therapy, as needed, has had a consistent beneficial effect on the patient. Some animals enter a complete remission and most animals have an improvement of seventy-five percent or more in their existing condition. To date, we have animals that have been as long as three years on this regimen whereby a holistic approach to overall health is combined with the minimal amount of drug therapy to extend the clinical remission and enhance their well-being and longevity. Finally, an important component of the program requires the regular monitoring of the pet with laboratory profiles to establish that health and endocrine balance is maintained. The frequency of this monitoring can be as often as every month or two, but is usually every six months to one year. This is particularly important with geriatric animals so that changes in their health status are identified early in the course of the disorder so that intervention can be practiced. We encourage readers to try this approach to the management of their patients. It is easy to implement, employs a common sense methodology, and is inexpensive. The clients and patients like it and, best of all, it works!

from: Dodds. W.J. "1990 Update on Autoimmune Diseases of Purebred Dogs". AKC Gazette Vol. 107, #7, July 1990, page 92.

Clinical Signs of Canine Hypothyroidism

Alterations in cellular metabolism
- -lethargy
- -mental dullness
- -exercise intolerance
- -weight gain
- -cold intolerance
- -mood swings or hyperexcitability
- -neurologic signs (including polyneuropathy and seizures)

Reproductive disorders
- -infertility of either sex
- -lack of libido
- -testicular atrophy
- -hypospermia
- -aspermia
- -prolonged interestrus interval
- -failure to have heat cycles
- -weak, dying or stillborn pups
- -enlarged mammary glands or lactation in non-pregnant bitches (pseudopregnancy)

Neuromuscular problems
- -weakness
- -stiffness
- -laryngeal paralysis
- -knuckling or dragging feet
- -muscle wasting

Dermatologic diseases
- -dry, scaly skin
- -coarse, dull coat
- -bilaterally symmetrical hair loss
- -"rat tail"
- -"puppy coat"
- -hyperpigmentation
- -seborrhea
- -pyoderma
- -myxedema

Cardiac abnormalities
- -bradycardia or slow heart rate
- -cardiac arrythmias
- -cardiomyopathy

Ocular diseases
- -corneal lipid deposits
- -corneal ulceration
- -uveitis
- -kerato-conjunctivitis sicca or "dry eye"

Gastrointestinal disorders
- -constipation
- -diarrhea
- -vomiting

Hematologic disorders
- -bone marrow failure
- -bleeding

—WJD

from: Dodds, W.J. "1990 Update on Autoimmune Diseases of Purebred Dogs". AKC Gazette Vol. 107 #7, July 1990, page 88.

Factors Associated With Autoimmune Disease

Sex (females 2:1 over males)
Genetic or familial history
Increasing frequency
Pregnancy
Nutritional influences
- -imbalance of trace dietary elements
- -preservatives or additives
- -food allergies to grains or meats
- -deficiencies of trace elements

Drug reactions
- -trimethoprim-sulfas
- -nitrofurans
- -butazolidin
- -phenobarbitol
- -primidone
- -diethylcarbamazine-oxybendazole
- -ivermectin

Stress
- -environmental
- -emotional
- -physiological

Hormonal irregularities
- -polyglandular autoimmunity (endocrinopathy)
- -pituitary-thyroid axis dysfunction
- -reproductive failure (abnormal heat cycles, pyometra, false pregnancy, hypogonadism, oligospermia, aspermia, anestrus)

Underlying or concomitant disease
- -lymphoma or leukemia (retrovirus infections)
- -bone marrow failure
- -immune dysregulation (humoral or cellular immunodeficiency)

Virus infection
- -parvoviruses
- -retroviruses
- -cytomegalovirus
- -measles and distemper viruses
- -hepatitis viruses

Frequent or recent use of modified live virus vaccines
- -parvovirus, -distemper, -rabies — vaccines alone or in combination

Other autoimmune disorders
- -Hashimoto's thyroiditis
- -Addison's disease
- -rheumatoid arthritis
- -lupus erythematosus
- -idiopathic thrombocytopenic purpura
- -hemolytic anemia
- -chronic active hepatitis
- -diabetes mellitus
- -hypogonadism
- -myasthenia gravis
- -pemphigus, vitiligo
- -glomerulonephritis
- -alopecia
- -Graves' disease
- -hypoparathyroidism
- -seizures
- -uveitis

—WJD

GENETIC ILLNESS: THE PUREBRED DILEMMA*

W. Jean Dodds, DVM
HEMOPET
938 Stanford Street
Santa Monica, CA 90403
(310) 828-4804
FAX (310) 828-8251

* Presented at AVAR Session, "Veterinary Medicine-Issues and Activism", 132nd AVMA Convention, July 10, 1995, Pittsburgh, PA.

AKC DELEGATE'S HIRIE COMMITTEE
(Preliminary Report, 1994)

Rank	Condition	Number of Breed Clubs +
1	Hypothyroidism	73
2	Arthritis	63
3	Allergies	62
4	Seizures	60
5	Cataracts	59
6	Hip dysplasia	58
7	Mammary cancer	55
8	Pyometra	52
9	Infertility	50
10	Cesarean section	50

+ Identified as fairly or very important (total number of responding clubs = 90).

OFA FREQUENCY OF HIP DYSPLASIA*
(Dogs born ≤ 1980 vs 1991-92)

Breeds with Frequency Reduced by > 50%		Breeds with No Change or Increased Frequency
Australian Shepherd	German Wirehaired Pointer	Afghan Hound
Bearded Collie	Old English Sheepdog	Boykin Spaniel
Bernese Mountain Dog	Portuguese Water Dog	Flat Coated Retriever
Bichon Frise	Puli	Kerry Blue Terrier
Borzoi	Rhodesian Ridgeback	Komondor
Brittany Spaniel	Siberian Husky	
Doberman Pinscher	Soft Coated Wheaten Terrier	
English Cocker Spaniel	Welsh Springer Spaniel	
English Setter		

* Corley, EA, Keller, G. Orthopedic Foundation for Animals, 1995, 9 pp. Total number tested 50,428 (71 breeds). Frequency 17.4% (≤ 1980) vs 12.4% (1991-92) [p=<0.05].

HEALTH PROBLEMS IN OLD ENGLISH SHEEPDOGS - 1990*

Rank	Condition	Number and % (n=740)
1	Diarrhea	142 (19%)
2	Sebaceous cysts	135 (18)
3	Arthritis	115 (16)
4	Hip dysplasia	113 (15)
5	Excitability	100 (14)
6	Hypothyroidism	99 (13)
7	Ear infection	97 (13)
8	Flea allergy	95 (13)
9	Pseudopregnancy +	51 (11)
10	Flatulence	76 (10)

* Dodds, WJ, Ward, PD, Velasquez, GR. Old English Times 12, 15-18, 1991; 2, 13-19, 1992.

+ All female (n=453).

HEALTH PROBLEMS IN OES BY DISEASE CATEGORY - 1990*

Rank	Disease category	Number and % (n= 740)
1	Thyroid disease & related conditions	357 (48%)
2	Skin & coat disorders	279 (38)
3	Gastrointestinal disorders	237 (32)
4	Joint & bone disorders	217 (29)
5	Temperament & behavior problems	214 (29)
6	Reproductive disorders	211 (29)
7	Chronic infections	182 (25)
8	Endocrine diseases	138 (19)
9	Thyroid diseases	129 (17)
10	Hip dysplasia	113 (15)

* Dodds, WJ, Ward, PD, Velasquez, GR. Old English Times 12, 15-18, 1991; 2, 13-19, 1992.

HEALTH PROBLEMS IN NEWFOUNDLANDS - 1989*

Rank	Condition	Number and % (n=1853)	
1	Hip dysplasia	392	(21%)
2	Dermatitis, allergic or nonspecific	272	(15)
3	Hypothyroidism	131	(7)
4	Entropion	100	(5)
5	Acute infectious diseases	72	(4)
6	Ruptured cruciate ligament	69	(4)
7	Trembling	66	(4)
8	Undershot jaw	64	(4)
9	Cardiac diseases including subaortic stenosis	60	(3)
10	Panosteitis	46	(3)

* Stanevich, R. Newfoundland Club of America Health and Longevity Committee, 1989, 32 pp.

HEALTH PROBLEMS IN BEARDED COLLIES - 1992*

Rank	Condition	Number and % (n=800)	
1	Diarrhea	154	(19%)
2	Flea allergy	140	(18)
3	Sebaceous cysts	119	(15)
4	Food hypersensitivity or intolerance	101	(13)
5	Fearfulness	89	(11)
6	Umbilical hernia	79	(10)
7	Shyness	69	(9)
8	Poor, dry, or thin coat	68	(9)
9	Hot spots	68	(9)
10	Chewing skin or coat	65	(8)

* Aronson, LP. Beardie Bulletin 22(3), 7-10, 1992; 24 (1 and 4), 7-11, 1994 and 1995.

HEALTH PROBLEMS IN IRISH WOLFHOUNDS - 1992*

Rank	Condition	Number and % (n=179)	
1	Presumed healthy	118	(66%)
2	All health problems	72	(40)
3	Chronic infections	48	(27)
4	Reproductive disorders	28	(16)
5	Allergies	22	(12)
6	Haircoat or pigment change	13	(7)
7	Diarrhea	8	(4)
8	Thyroid diseases	7	(4)
9	Arthritis	7	(4)
10	All other conditions	24	(13)

* Dodds, WJ. Harp and Hound 2, 28-40, 1992.

HEALTH PROBLEMS IN IRISH SETTERS - 1992*

Rank	Condition	Number and % + (n=356)	
1	Hypothyroidism	163	(46%)
2	Cancers	154	(43)
3	Seizures	135	(38)
4	Hip dysplasia	127	(36)
5	Panosteitis	83	(23)
6	Spondylosis	82	(23)
7	Heart diseases	73	(20)
8	Hypertrophic osteodystrophy	65	(18)
9	Cataracts	61	(17)
10	Autoimmune diseases	52	(15)

* Brown, SG. Irish Setter Club of America and Institute Genetic Disease Control, 1992, 31 pp.

+ Number of responses.

IMPORTANCE OF GENETIC SCREENING FOR HEALTH AND LONGEVITY OF PUREBRED DOGS

W. Jean Dodds, DVM*
Wadsworth Center
NYS Department of Health
Albany, NY 12201-0509

The common practice to linebreed and inbreed purebred dogs facilitates the transmission and recognition of genetic defects. An effective mechanism to discover and eventually control the frequency of these defects is to develop large-scale screening programs for the identification of affected and/or carrier animals within the population at large. Screening programs of this type have been used successfully in humans for many years (e.g., for Tay-Sachs disease, phenylketonuria) and more recently have been applied to animals (e.g., for mannosidosis in cattle; hip dysplasia, eye and blood diseases in dogs).

In the mid-1960s veterinarians began screening dog populations for inherited eye diseases and hip dysplasia. Large-scale screening programs for companion animals are an effective means of genetic control because they live in daily contact with their owners, and thus illnesses are more likely to be noticed and treated. Genetic screening may be essential to the survival of breeds in which mild or moderately severe defects have been propagated unknowingly for many generations.

Depending on the mode of inheritance, different approaches need to be applied for the detection and control of genetic diseases. It is advantageous to be able to select against heterozygotes (carriers) rather than to have to eliminate affected individuals from a breeding program once the condition is manifested. Control and elimination of the disease by testing are feasible and reliable in cases where the asymptomatic or carrier state has an expressed biochemical marker. Some current examples include testing for von Willebrand's disease and hemophilia, autoimmune thyroid disease, the various metabolic enzyme and storage disorders, and neuromuscular diseases. Coupled with this approach to eliminating undesirable traits is the necessity to monitor breeding stock regularly for overall soundness, reproductive health and performance.

With respect to the increasing frequency of thyroid and other autoimmune diseases amongst purebreds (see July 1990 AKC Gazette article), a complete thyroid profile including thyroid autoantibody tests can be used to evaluate the fitness of healthy stock for breeding. A bitch with antithyroid antibodies in her blood may pass these along to her puppies with the colostral milk. Also, any dog having antithyroid antibodies is at potential risk to develop other autoimmune diseases.

Thyroid testing for genetic screening purposes is less likely to be meaningful before puberty. Healthy young dogs (less than 15 months of age) should have thyroid baseline levels for all parameters in the upper third of the adult normal ranges. In fact, for optimum function, thyroid levels of adults should be at least at mid-range for breeding and performance. Thyroid therapy should be given to correct any thyroid metabolic imbalance, reduce the risk of developing other related immune-mediated disorders, and to control or prevent the process of thyroiditis from progressing to depletion and exhaustion of the thyroid gland (which is expressed as the clinical syndrome of hypothyroidism).

It is important that the top-producing sires and foundation bitches of a breed be screened for these conditions because they represent the major nucleus of genetic material for the current and future decades. Despite the proven benefits of this approach and intensive educational efforts, it should be recognized that some breeders choose not to participate. Veterinarians and scientists who provide testing and counseling services should be understanding in dealing with the concerns and sensitivities of these individuals. Attempts to pressure people into participation can be counterproductive to the overall goals of the program. Success depends upon an honest, voluntary commitment to the principles and ethics of breeding better companion animals.

---

* Home Address: Hemopet, 938 Stanford Street, Santa Monica, CA 90403 (213-828-4804)

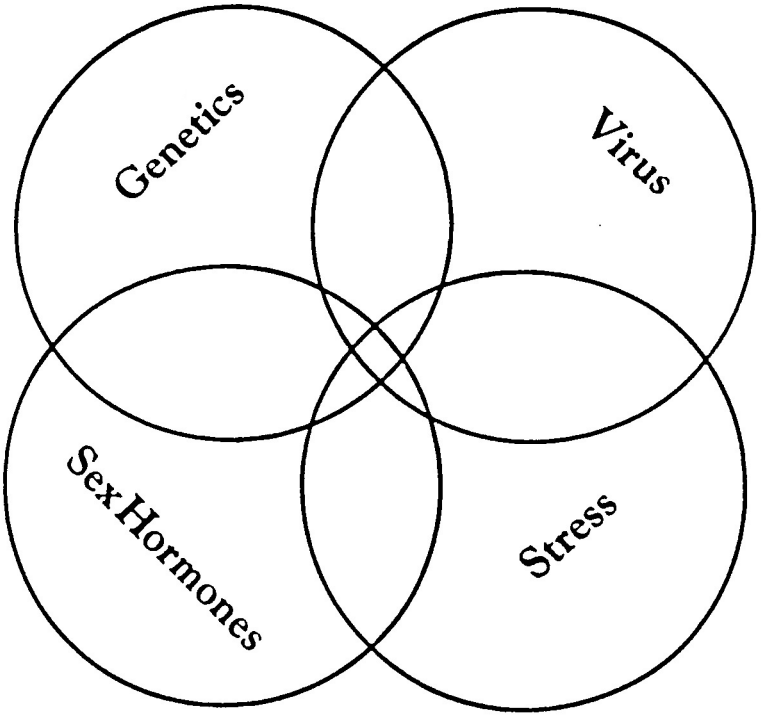


Fig 1. Multifactorial etiology of autoimmune diseases.

from Talal, N. Cyclosporine as an Immunosuppressive Agent for Auto immune Disease: Theoretical Concepts and Therapeutic Strategies. Transpl. Proc. Vol. 20 Supplement 4 June 1988

MORE BUMPS ON THE VACCINE ROAD: ISSUES OF EFFICACY AND SAFETY OF HUMAN AND ANIMAL VACCINES

W. Jean Dodds, DVM
HEMOPET
938 Stanford Street
Santa Monica, CA 90403
310-828-4804/FAX 310-828-8251

The challenge to develop effective and safe vaccines for some of the more prevalent infectious diseases of humans and animals has become increasingly difficult. Top vaccine researchers attribute this problem to the lack of world leadership and funding, and face tough scientific hurdles in developing future vaccines. Despite the urgent need to develop an effective vaccine for AIDS, the goal remains elusive. Recent concerns about vaccine safety and the increasing awareness of the potential for adverse vaccine reactions have culminated in a moratorium on the use of oral polio vaccine, and a questioning of the overall safety of childhood vaccines such as those for pertussis and measles.

In veterinary medicine, evidence implicating vaccines in triggering immune-mediated and other chronic disorders (vaccinosis) is growing. While some of these problems have been traced to contaminated or poorly attenuated batches of vaccine that revert to virulence, others reflect the host's genetic predisposition to adverse vaccinal events following use of the monovalent or polyvalent products given routinely to animals.

Determining causality for adverse effects of vaccines is based on three questions: Can it ? (potential causality); Did it ? (retrodictive causality); and Will it? (predictive causality). Other factors to be considered include: vaccine dosage in relation to body mass and age; advantages and disadvantages of modified-live (attenuated) and killed (inactivated) vaccines; hormonal state during vaccination; and periodicity of booster vaccinations. Alterative to current vaccinology include: measuring serum antibody titers; avoidance of unnecessary vaccines or overvaccinating; caution in vaccinating sick, very old, debilitated, or febrile individuals, and families known to be at high risk for immunologic reactions; and homeopathic nosodes. A multifaceted approach is needed to further recognition of this situation, along with implementing alternative strategies to contain infectious diseases and reduce the environmental impact of conventional vaccines.

ADVERSE EFFECTS OF VACCINES*

Causality Questions

- **Can it?** (Potential Causality)
  - Strength of association
  - Analytic bias
  - Biologic gradient (dose-reponse effects)
  - Statistical significance
  - Consistency
  - Biologic plausibility and coherence
- **Did it?** (Retrodictive Causality) *(Cause) Review history*
  - General experience with the vaccine
  - Alternative etiologic candidates
  - Susceptibility of vaccine recipient
  - Timing of events *
  - Characteristics of adverse events *
  - Dechallenge
  - Rechallenge *
- **Will it?** (Predictive Causality)
  - Magnitude of risk difference
  - Attributable risk
  - Depends on **Can it?**
  - Difficult to quantitate

---

* Adapted from Stratton KR, Howe CJ, Johnston Jr RB (eds). Adverse Events Associated with Childhood Vaccines. Evidence Bearing on Causality. Nat'l Acad. Press, Washington DC, 1994.

Dr. Sholtz vaccine info. - Wisconsin

Partial dosage - sign release for vet.

VACCINE DOSAGE

Body Mass

20-30 & under

- Same dose intended for toy and giant breeds
- Why?
- MLV vaccines-- immunogenic principle not based on body mass
- Killed vaccines -- should be adjusted for body mass –
- Minimum/optimum doses for protection
- Excess antigen present

stimulates the immune system

Age

- Optimal age for response
  - 12 weeks + for puppies
  - 10 weeks + for kittens
  - same for all breeds and sizes?
- Earliest age to promote safety
  - 6 weeks for puppies and kittens
- Effective age varies
- Blocking effects of maternal immunity

- At 22 weeks of age, if the immune system is OK, Dr, Dodds & Dr Sholtz feel vaccines wouldn't be required.

KILLED (INACTIVATED) VACCINES

Advantages

- No residual virulence
- No reversion to virulence
- Cannot multiply in host
- Less likely to cause immunosuppression
- Less delayed type vaccinosis

Disadvantages

- Shorter duration of immunity
- Less cellular or secretory immunity
- Adjuvant required
- More boosters needed
- Hypersensitivity more likely
- More costly to produce

HORMONAL STATE DURING VACCINATION

Avoid Vaccinating Pet Animals

- Period (30 days) before estus
- During or immediately after estrus

re: modified live vaccines

- During pregnancy
- Post-partum (lactation)

PERIODICITY OF BOOSTER VACCINATIONS

Public Health Issue

*** • No evidence that annual boosters are necessary

- Need to lengthen interval between boosters (e.g. every 2-3 years for healthy adults)

* • Geriatric animals vaccinated with caution especially if chronically diseased *immune system decreasing*

- Consider monitoring serum antibody titers instead

1. secretions
2. tissues
3. blood values

MODIFIED-LIVE (ATTENUATED) VACCINES

Advantages

- Longer duration of immunity
- More complete immunity
- Adjuvant not required
- Fewer boosters needed
- Less costly to produce
- Hypersensitivity less likely

Disadvantages

- Residual virulence
- Reversion to virulence
- Multiply in host (viremic phase) *Day 3 for 10 days after.*
* • Immunosuppression
- Vaccinosis of delayed type

ALTERNATIVES TO CONVENTIONAL VACCINATION

Measure Serum Anitbody Titers - *measures IgE*

- Annual titers against common infectious agents
- Offer as alternative to routine boosters - *titers can be done every year, test for Parvo, Distemper.*
- Use for chronically ill, allergic, geriatric, or immunocompromized animals

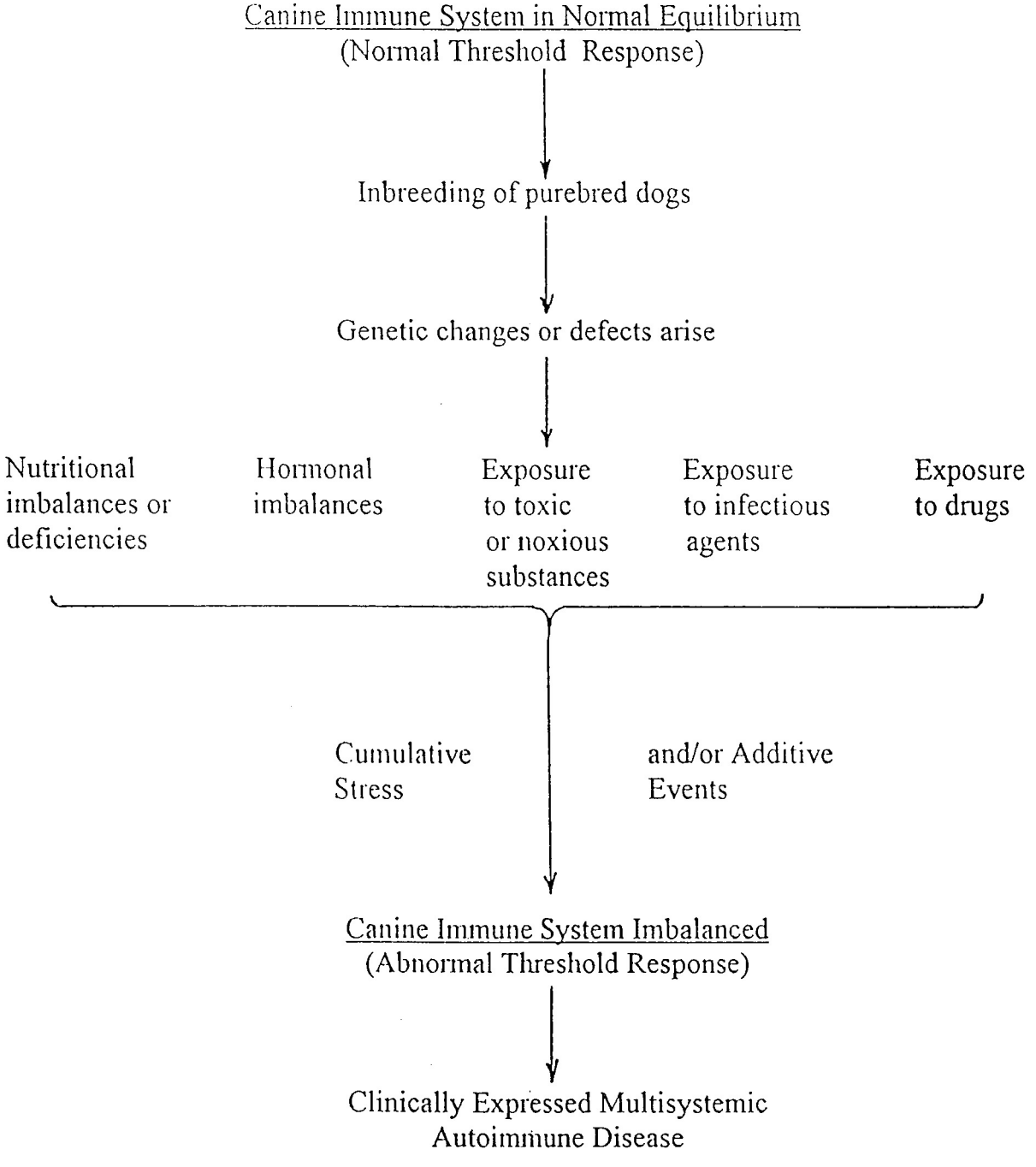
Avoid Vaccinating or Overvaccinating

- Sick, very old, debilitated animals
- Immunocompromized animals
- Febrile animals
- Certain breeds/families (e.g. Akita, Weimaraner, harlequin Great Dane)

Alternative Methodologies

- Need informed consent and doctor/client/ patient relationship
- Homeopathic alternatives (nosodes), while safe, need controlled trials to establish efficacy

THE THRESHOLD MODEL FOR INDUCING AUTOIMMUNE DISEASE



RELATED AKITAS WITH VACCINE-ASSOCIATED DISEASE

No. Dogs	Sex	Cause of death (age 5 mo -2 yrs)	Age at onset (weeks)	Time post-vaccines (days)	Cyclic episodes of pain	High spiking fevers	Poly - arthritis
8	3M	1heartworm disease	Mean age	Mean reaction time	6/8	8/8	5/8
(3 sets of litter-mates)	5F	1 renal dz 1 sepsis 1 DIC 3 euthanized 1 unknown	14 Median age 15½	14			

(Liver)

No. Dogs	Alk Phos ↑	ALT ↑	AST ↑	CPK ↑	BUN ↑	Joint aspirate/ culture
8	6/8	3/5	2/4	1/2	2/4	2 nonseptic purulent *white blood cells*
						1 nonseptic lymphocytic

⇧ *no bacteria present: vaccine probable cause*

[5 tested neg. for Borrelia , RMSF, Ehrlichia , Brucella , ANA , LE-prep , RF , direct Coombs , blood culture. 1 ANA = 1:40]

white cells *platlets*

No. Dogs	Parasites	Anemia	Leuko-cytosis	Neutrophilia	Thrombo-cytopenia
8	2/3	5/5	7/7	5/6	2/6

[2 necropsied: 1 glomerular amyloidosis, myocarditis, endocarditis, enteritis, pyelonephritis. 1 eosinophilic enteritis]

WEIMARANERS WITH VACCINE-ASSOCIATED DISEASE

No. Dogs	Sex	Age at onset (weeks)	Time post-vaccines (days)	Cyclic episodes of pain	High spiking fevers	Poly-arthritis (HOD)
24	13 M 5F 6 unknown	8-20 Mean age 13½	1-27 Mean reaction time 10½	+ (all)	+ (all)	+ (all)

Leuko-cyctosis	Diarrhea	Lethargy	Anorexia	Enlarged lymph nodes	IgA , IgG , IgM level	Outcome
+	+	+	+	variable	variable	2 died 3 euthanised 12 healthy adults 3 sick adults 4 unknown

REVISED VACCINATION PROTOCOL - 1996

W. Jean Dodds, DVM
HEMOPET
938 Stanford Street
Santa Monica, CA 90403

Note: This schedule is the one I recommend and should **not** be interpreted to mean that other protocols recommended by a veterinarian would be less satisfactory. It's a matter of professional judgement and choice.

For breeds or families of dogs susceptible to or affected with immune dysfunction, immune-mediated disease, immune-reactions associated with vaccinations, or autoimmune endocrine disease (e.g., thyroiditis, Addison's or Cushing's disease, diabetes, etc.), the following protocol is recommended:

Age of Pups	Vaccine Type
6 weeks (range could be 5 ½ - 6 ½ weeks but not earlier)	Distemper + measles (*without* hepatitis)
6-8 weeks and 10 weeks	Distemper ± parainfluenza (*without* hepatitis) for pups **not** receiving measles earlier
7-8 weeks and } same schedule and 10-11 weeks } product each time	Killed canine parvovirus * given 3-4 weeks earlier
12 weeks	Distemper+hepatitis+parainfluenza ± leptospirosis (*without* parvovirus)*
14 weeks and } same schedule and 18-20 weeks } product each time**	Distemper+hepatitis+parainfluenza ± leptospirosis + killed canine parvovirus *
16-24 weeks	Killed rabies vaccine

* During parvovirus epidemics or for highly susceptible breeds such as Rottweilers, newer modified-live virus (MLV) vaccines that override maternal immunity may be needed here.

** Alternatively, killed parvovirus vaccine * and the distemper + hepatitis + parainfluenza ± leptospirosis combination are alternated every 2 weeks during the 14-20 week period.

- **Annual boosters** using distemper + hepatitis + parainfluenza ± leptospirosis + killed canine or MLV parvovirus are given to adults until mid-life. Thereafter, boosters are given every 2-3 years until old age. Beyond 10 years of age, booster vaccinations are generally not needed, and may be unwise if aging or other diseases are present. For animals at high exposure risk to parvovirus disease, an additional parvovirus vaccination can be given at the 6-month point.

- I use only killed 3-year rabies vaccine for adults and give it separated from other vaccines by at least 2 and preferably 3-4 weeks.

- I do **not** use Bordetella or corona virus vaccines unless these diseases are endemic in the local area or specific kennel. I do **not** recommend Lyme vaccine.

- I do **not** recommend vaccinating bitches during estrus, pregnancy or lactation.

- I recommend that distemper or distemper-measles vaccine be given *without* hepatitis before the age of 10 weeks, because of the reported suppression of lymphocyte responsiveness induced by polyvalent canine distemper and adenovirus vaccines (Phillips et al., Can J Vet Res 1989; 53: 154-160.)

- For animals previously experiencing adverse vaccine reactions or breeds at higher risk for such reactions (e.g. Weimaraners, Akitas), **alternatives to booster vaccinations** should be considered. These include avoiding boosters except rabies vaccine as required by law; annually measuring serum antibody titers against specific canine infectious agents, such as distemper and parvovirus; and homeopathic nosodes. [This last option is considered an unconventional treatment that has not been scientifically proven to be efficacious. However, data from Europe and clinical experience in North America support its use, and controlled studies are underway to test the method under challenge conditions. If veterinarians choose to use homeopathic nosodes, their clients should be provided with an appropriate disclaimer and written informed consent should be obtained.]

PET LIFE-LINE **HEMOPET**

W. Jean Dodds

W. Jean Dodds, DVM
310-828-4804
FAX 310-828-8251

HEALTH MEDICINE

THE IMMUNE SYSTEM: *guardian against disease*

Dysfunction in the immune system can compromise the body's entire line of defense

By W. Jean Dodds, D.V.M.

The immune system plays a central role in maintaining the body's overall general health and resistance to disease. This article focuses on environmental factors or events that may cause or trigger immune dysfunction leading to either immune deficiency or immune stimulation (reactive or autoimmunity). Related to these events is the development of cancer, which is a disruption of cell growth control. [For further information about the immune system, see the following issues of DOG WORLD: April 1992, p. 36; May 1992, p. 44; and October 1992, p. 40.]

Immune competence is provided and maintained by two cellular systems that involve lymphocytes. Lymphocytes are cells produced by the body's primary (bone marrow and thymus) and secondary (lymph nodes and spleen) lymphatic organs. They are descendants of the bone marrow's pool of stem cells, and produce a circulating or humoral immune system derived from B-cells (bursa-dependent or bone marrow-derived), and a cellular or cell-mediated immune system that derives from T-cells (thymus-dependent).

B-cell immunity includes the circulating antibodies or immunoglobulins such as IgG, IgM, IgA, IgD and IgE. These antibodies provide an important defense mechanism against disease in healthy individuals but can become hyperactive or hypoactive in a variety of disease states. Hyperactive or increased levels of immunoglobulins can occur in two ways:

- acutely, as a reaction to disease or inflammatory insult ("acute-phase" reaction); or
- chronically, as in autoimmune or immune-mediated diseases, chronic infections and certain types of bone marrow and organ cancers.

Hypoactive or decreased levels of immunoglobulins can result from rare genetically based immunodeficiency states such as agammaglobulinemia or hypogammaglobulinemia, and from the immune suppression associated with chronic viral, bacterial or parasitic infections, cancers, aging, malnutrition, drugs, toxins, pregnancy, lactation and stress.

T-cell or cell-mediated immunity is the cellular mechanism whereby T-cells act as coordinators and effectors of the immune system. Cell-mediated immunity involves the lymph nodes, thymus, spleen, intestine (gut-associated lymphoid tissue), tonsils and a mucosal secretory immunity conveyed by IgA. The major classes of T-cells are designated as helper, cytotoxic and suppressor cells. The helper cells "help" coordinate the immune response, whereas the cytotoxic cells constitute the effector network that participates in removing virus-infected cells from the body. The third class of suppressor T-cells is important in dampening the immune response when it becomes overactive or out of regulatory control. Finally, cooperation between the various T-cell classes and between T- and B-cells is an important component of the normal humoral and cellular immune response.

Hyperactive cellular immune responses produce autoimmune and other immune-mediated diseases, while hypoactive cell-mediated immunity causes immune suppression and incompetence. Classical examples of this latter situation occur with retroviral infection such as human AIDS or the animal equivalents (e.g., feline immunodeficiency virus, feline leukemia virus, bovine leukemia virus, equine infectious anemia).

The term "autoimmunity" literally means immunity against self and is caused by an immune-mediated reaction to self-antigens (i.e., failure of self-tolerance). Susceptibility to autoimmune disease has a genetic basis in humans and animals. Numerous viruses, bacteria, chemicals, toxins and drugs have been implicated as the triggering environmental agents in susceptible individuals. This mechanism operates by a process of molecular mimicry and/or non-specific inflammation. The resultant autoimmune diseases reflect the sum of the genetic and environmental factors involved. Autoimmunity is most often mediated by T-cells or their dysfunction. As stated in a review, "perhaps the biggest challenge in the future will be the search for the environmental events that trigger self-reactivity." (Sinha, Lopez and McDevitt; *Science*, 248: 1380, 1990.)

The four main causative factors of autoimmune disease have been stated to be:

- Genetic predisposition
- Hormonal influences, especially of sex hormones
- Infections, especially of viruses
- Stress

Immune-suppressant viruses of the retrovirus and parvovirus classes have recently been implicated as causes of bone marrow failure, immune-mediated blood diseases, hematologic malignancies (lymphoma and leukemia), dysregulation of humoral and cell-mediated immunity, organ failure (liver, kidney) and autoimmune endocrine disorders—especially of the thyroid gland (thyroiditis), adrenal gland (Addison's disease) and pancreas (diabetes). Viral disease and recent vacci-

mits treatment intervention, where indicated, to avoid the appearance or advancement of clinical signs associated with hypothyroidism. For optimal health, dogs younger than 15 to 18 months of age should have thyroid baseline levels in the upper half of the adult normal ranges. This is because puppies and adolescent dogs require higher levels of thyroid hormones as they are still growing and maturing. Similarly, older animals beyond 8 or 9 years of age have slower metabolisms and so baseline thyroid levels of normal (euthyroid) dogs may be slightly below midrange. For optimum thyroid function of breeding stock, levels should be close to the midpoint of the laboratory normal ranges, because lower levels may be indicative of the early stages of thyroiditis among relatives of dog families previously documented to have thyroid disease.

The difficulty in accurately diagnosing early thyroid disease is compounded by the fact that some patients with typical clinical signs of hypothyroidism have circulating thyroid levels within the normal range. A significant number of these patients will improve clinically when given thyroid medication. In such cases, blood levels of the hormones can be normal but tissue levels are inadequate to maintain health, and so the patient shows clinical signs of hypothyroidism. This situation pertains in selenium deficiency (discussed below). While animals in this category should respond well to thyroid medication, only experienced clinicians are likely to recognize the need to place these dogs on a six- to eight-week clinical trial of thyroid supplementation. This approach is safe and clinically appropriate, but it requires rechecking blood levels of thyroid hormones toward the end of the six- to eight-week period to assure that the patient is receiving the correct dose of medication.

Because animals with autoimmune thyroid disease have generalized metabolic imbalance and often have associated immunological dysfunction, it is advisable to minimize their exposures to unnecessary drugs, toxins and chemicals and to optimize their nutritional status with healthy balanced diets. Wholesome nutrition is a key component of maintaining a healthy immune system. In our experience, families of dogs susceptible to thyroid and other autoimmune diseases show generalized improvement in health and vigor when fed premium cereal-based diets preserved naturally with vitamins E and C without the addition of chemical antioxidant preservatives such as BHA, BHT or ethoxyquin. Fresh home-cooked vegetables with herbs, low-fat dairy products and meats such as lamb, chicken and turkey can be added as supplements. Challenging the immune system of animals susceptible to these disorders with polyvalent modified-live vaccines has been associated with adverse effects in some cases (see below). Certain drugs should be avoided whenever possible in susceptible or affected animals. These include: potentiated sulfonamide antibiotics (trimethoprim- and ormetoprim-sulfas); nitrofuran antibiotics; anticonvulsants (phenobarbital, primidone); anti-inflammatory drugs (phenybutazone); monthly or combination heartworm preventives (ivermectin, milbemycin oxime, diethylcarbamazine-oxybendazole).

Nutritional influences can have a profound effect on thyroid metabolism. For example, iodine deficiency in areas where cereal grain crops are grown on iodine-deficient soil will impair thyroid metabolism because this mineral is essential for formation of thyroid hormones. Recently an important link has been shown between selenium deficiency and hypothyroidism. Selenium is an important mineral for maintaining muscle strength and function, and along with vitamin E is needed for a healthy immune system. Cereal grain crops grown on selenium-deficient soil will contain relatively low levels of selenium. While commercial pet food manufacturers compensate for variations in basal ingredients by adding vitamin and mineral supplements, it is difficult to determine optimum levels for so many different breeds of dogs having varying genetic backgrounds and metabolic needs.

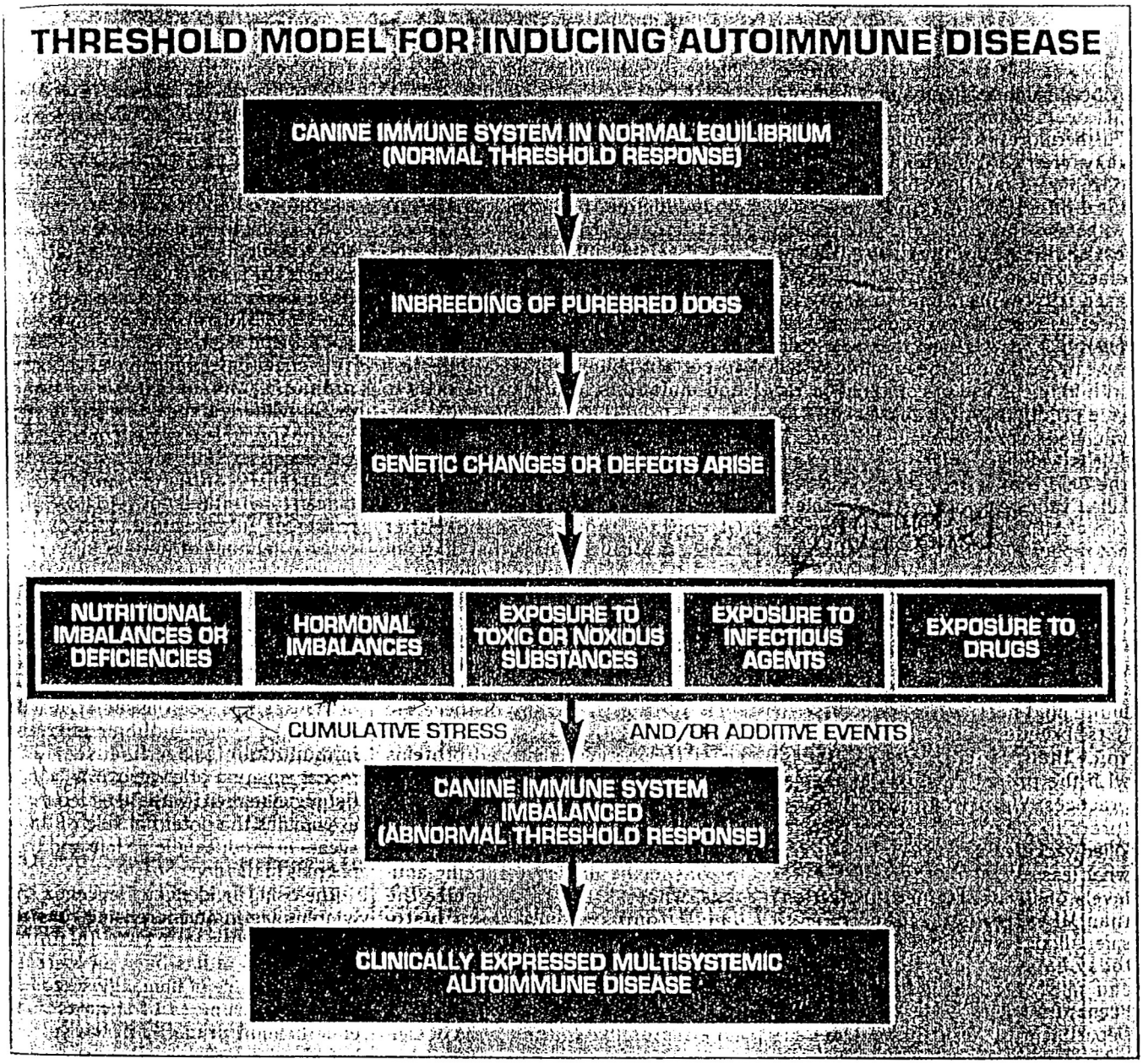
The selenium-thyroid connection has significant clinical relevance, because blood levels of both bound and unbound (free) T4 rise in selenium deficiency. However, this effect does not get transmitted to the tissues because contrary to the expected central regulatory response to these increased T4 levels, blood levels of thyroid-stimulating hormone (TSH) do not go down but are elevated or unchanged. Thus, selenium-deficient individuals showing clinical signs of hypothyroidism could be overlooked on the basis that blood levels of T4 hormones appeared normal. The selenium issue is further complicated because chemical antioxidants (such as BHA, BHT or ethoxyquin) can impair the bioavailability of vitamin A, vitamin E and selenium and alter cellular metabolism by inducing or lowering cytochrome p-450, glutathione peroxidase (a selenium-dependent enzyme) and prostaglandin levels. The way to avoid this problem is to use foods preserved with natural antioxidants such as vitamin E and vitamin C.

IMMUNOLOGICAL EFFECTS OF VACCINES

Combining viral antigens, especially those of modified-live virus (MLV) type which are attenuated to prevent reversion to virulence but still multiply in the host, elicits a stronger antigenic challenge to the animal. This is in contrast to killed virus vaccines which are inactivated and cannot multiply in the tissues of the vaccinated animal or person. The amplification of MLV vaccines by multiplication is often viewed as desirable because a more potent immunogen presumably mounts a more effective and sustained immune response. However, it can also overwhelm the immunocompromised or even a healthy host that is continually bombarded with other environmental stimuli and has a genetic predisposition that promotes adverse response to viral challenge. This scenario may have a significant effect on the recently weaned young puppy that is placed in a new environment. Furthermore, while the frequency of vaccinations is usually spaced two to three weeks apart, some veterinarians have advocated vaccination once a week in stressful situations.

To me, this practice makes no sense from a scientific or medical perspective. While young puppies exposed this frequently to vaccine antigens may not demonstrate overt adverse effects, their relatively immature immune systems may be temporarily or more permanently harmed from such antigenic challenges. Consequences in later life may be the increased susceptibility to chronic debilitating diseases. Some veterinarians trace the increasing current problems with allergic and immunological diseases to the introduction of MLV vaccines some 20 years ago. While other environmental factors no doubt have a contributing role, the introduction of these vaccine antigens and their environmental shedding may provide the final insult that exceeds the immunological tolerance threshold of some individuals in the pet population (Figure 1, p. 29).

Manufacturers of MLV combination vaccines recommend using the same dose for animals of all ages and different sizes. It has never made any sense to vaccinate toy and giant breed puppies (to choose two extremes) with the same vaccine dosage. While these products provide sufficient excess of antigen for the average-sized animal, it is likely to be either too much for the toy breeds or too little for the giant breeds. In addition, combining certain specific viral antigens such as distem-


THRESHOLD MODEL FOR INDUCING AUTOIMMUNE DISEASE
CANINE IMMUNE SYSTEM IN NORMAL EQUILIBRIUM (NORMAL THRESHOLD RESPONSE)
INBREEDING OF PUREBRED DOGS
GENETIC CHANGES OR DEFECTS ARISE
NUTRITIONAL IMBALANCES OR DEFICIENCIES
HORMONAL IMBALANCES
EXPOSURE TO TOXIC OR NOXIOUS SUBSTANCES
EXPOSURE TO INFECTIOUS AGENTS
EXPOSURE TO DRUGS
CUMULATIVE STRESS
AND/OR ADDITIVE EVENTS
CANINE IMMUNE SYSTEM IMBALANCED (ABNORMAL THRESHOLD RESPONSE)
CLINICALLY EXPRESSED MULTISYSTEMIC AUTOIMMUNE DISEASE

per with adenovirus 2 (hepatitis) has been shown to influence the immune system by reducing lymphocyte numbers and responsiveness.

Relatively little attention has been paid to the hormonal status of the patient at the time of vaccination. While veterinarians and vaccine manufacturers are aware of the general rule not to vaccinate animals during any period of illness, the same principle should apply to times of physiological hormonal change. This is particularly important because of the known role of hormonal change along with infectious agents in triggering autoimmune disease. Therefore, vaccinating animals at the beginning, during or immediately after an estrus cycle is unwise, as would be vaccinating animals during pregnancy or lactation. In this latter situation, adverse effects can accrue not only to the dam but also because a newborn litter is exposed to shed vaccine virus. One can even question the wisdom of using MLV vaccines on adult animals in the same household because of exposure of the mother and her litter to shed virus. Recent studies with MLV herpes virus vaccines in cattle have shown them to induce necrotic changes in the ovaries of heifers that were vaccinated during estrus. The vaccine strain of this virus was also isolated from control heifers that apparently became exposed by sharing the same pasture with the vaccinates. Furthermore, vaccine strains of these viral agents are known to be causes of abortion and infertility following herd vaccination programs. If one extrapolates these findings from cattle to the dog, the implications are obvious.

Most single and combination canine vaccines available today are of MLV origin. This is based primarily on economic reasons and the belief that they produce more sustained protection. A long-standing question remains, however, concerning the comparative safety and efficacy of MLV vs. killed (inactivated) virus vaccines. A recent examination of the risks posed by MLV vaccines concluded that they are intrinsically more hazardous than inactivated products. The residual virulence and environmental contamination resulting from the shedding of vaccine virus is a serious concern. More importantly, the ability of new infective agents to develop and spread poses a threat to both wild and domestic animal populations.

The controversy in weighing the risks and benefits of MLV vs. killed vaccines is building. Vaccine manufacturers seek to achieve minimal virulence (infectivity) while retaining maximal immunogenicity (protection). This desired balance may be relatively easy to achieve in clinically normal, healthy animals but may be problem-

atic for those with even minor immunologic deficit. The stress associated with weaning, transportation, surgery, subclinical illness and a new home can also compromise immune function. Furthermore, the common viral infections of dogs cause significant immunosuppression. Dogs harboring latent viral infections may not be able to withstand the additional immunological challenge induced by MLV vaccines. The increase in vaccine-associated distemper and parvovirus diseases are but two examples of this potential. So, why are we causing disease by weakening the immune system with frequent use of combination vaccine products? After all, vaccines are intended to protect against disease.

It is well-recognized by experts in the field that a properly constituted killed vaccine is always preferable to one of MLV origin. Killed vaccines do not replicate in the vaccinated animal, do not carry the risk of residual virulence and do not shed attenuated viruses into the environment. On the other hand, MLV vaccines are capable of stimulating a more sustained protective response. So what does the future hold here? Veterinarians, scientists, breeders and owners need to voice their concern and discontent with the present industrial vaccine practices. We need to urge manufacturers to seek alternatives. Even if killed vaccines are proven to be somewhat less efficacious (produce lower levels or less sustained protection) than MLV products, they are more safe. All killed vaccines on the market today have passed current efficacy and safety standards in order to be licensed for use by the United States Department of Agriculture. The issue is to what extent being more effective elicits a benefit rather than a risk.

In the future we will see the evolution of new approaches to vaccination including sub-unit vaccines, recombinant vaccines using DNA technology and killed products with new adjuvants to boost and prolong protection. These are not simple solutions to a problem, however, because early data from recombinant vaccines against some human and mouse viruses have shown potentially dangerous side effects by damaging T-lymphocytes. Contributing factors were shown to be the genetic background of the host, the time or dose of infection and the makeup of the vaccine. We are obviously still a long way from producing a new generation of improved and safe vaccines. In the meantime, we need to return to using killed products whenever they are available and should consider giving them more often (twice yearly rather than annually) for high-risk exposure situations. Vaccines, while necessary and generally safe and efficacious, can be harmful or ineffective in selected situations.

CANCER AND IMMUNITY

Proper regulation of cellular activity and metabolism is essential to normal body function. Cell division is a process under tight regulatory control. The essential difference between normal and tumor or cancerous cells is a loss of growth control over the process of cell division. This can result from various stimuli such as exposure to certain chemicals, viral infection and mutations, which cause cells to escape from the constraints that normally regulate cell division. Proliferation of a cell or group of cells in an uncontrolled fashion eventually gives rise to a growing tumor or neoplasm. Of course, tumors can be either benign (a localized mass that does not spread) or malignant (cancerous), in which the tumor grows and metastasizes to many distant sites via the blood or lymph.

Tumor cells also express a variety of proteins called "neoantigens" on their surface, and many of these are different to antigens found on normal cells. These new or altered proteins are recognized as foreign by the immune system and so trigger an immunological attack. There are a large number of them known as tumor-specific or tissue-specific antigens, whereas others recognize the blood group systems, major histocompatibility complex and viruses.

The situation in cancer is complex because not only can immunologically compromised individuals become more susceptible to the effects of cancer-producing viral agents and other chemical carcinogens, the cancer itself can be profoundly immunosuppressive. The form of immunosuppression usually varies with the tumor type. For example, lymphoid tumors (lymphomas and leukemia) tend to suppress antibody formation, whereas tumors of T-cell origin generally suppress cell-mediated immunity. In chemically induced tumors, immunosuppression is usually due to factors released from the tumor cells or associated tissues. The presence of actively growing tumor cells presents a severe protein drain on an individual that may also impair the immune response. Blocking factors present in the serum of affected animals exist that can cause enhancement of tumor growth. Additionally, immunosuppression in tumor-bearing animals can be due to the development of suppressor cells.

The body also contains a group of complimentary factors that provide a protective effect against tumors and other immunologic or inflammatory stresses. These are mixtures of proteins produced by T-cells and are referred to as "cytokines." Cytokines include the interleukins, interferons, tumor-necrosis factors and lymphocyte-derived growth factors. Recent studies have shown that normal levels of zinc are important to protect the body against the damaging effects of the specific cytokine, tumor-necrosis factor (TNF). Inadequate levels of zinc have been shown to promote the effect of TNF in disrupting the normal endothelial barrier of blood vessels. This could have a significant effect in promoting the metastasis of tumor cells to different sites, thereby hastening the spread and growth of a particular cancer.

Currently about 15 percent of human tumors are known to have viral causes or enhancement. Viruses also cause a number of tumors in animals and no doubt the number of viruses involved will increase as techniques to isolate them improve. The T-cell leukemias of humans and animals are examples of those associated with retroviral infections. This same class of viruses has been associated with the production of autoimmunity and immunodeficiency diseases. The recent isolation of a retrovirus from a German Shepherd with B-cell leukemia exemplifies the potential role of these agents in producing leukemia and lymphomas in the dog.

The rising incidence of leukemia and lymphomas in an increasing number of dog breeds is a case in point. Similarly, there has been an increase in the incidence of hemangiosarcomas (malignant tumors of the vascular endothelium) primarily in the spleen, but also in the heart, liver and skin. They occur most often in middle-age or older dogs of medium to large breeds. The German Shepherd Dog is the breed at highest risk, but other breeds including the Golden Retriever, Old English Sheepdog, Irish Setter and Vizsla have shown a significantly increased incidence especially in certain families. This suggests that genetic and environmental factors play a role. It is tempting to speculate that environmental factors that promote immune suppression or dysregulation contribute to failure of immune surveillance mechanisms. These protect the body against the infectious and environmental agents that induce carcinogenesis and neoplastic change.

NUTRITIONAL FACTORS AND THE IMMUNE SYSTEM

As alluded to previously, an adequate nutritional state is important in managing a variety of inherited and

other metabolic diseases as well as for a healthy immune system. Examples where nutritional management is important in inherited disorders include: adding ingredients to the diet to make it more alkaline for Miniature Schnauzers with calcium oxalate bladder or kidney stones; use of the vitamin A derivative, etretinate, in Cocker Spaniels and other breeds with idiopathic seborrhea of the skin; management with drugs and diet of diseases such as diabetes mellitus and the copper-storage disease prevalent in breeds such as the Bedlington Terrier, West Highland White Terrier and Doberman Pinscher; wheat-sensitive enteropathy (bowl disease) in Irish Setters; and treatment of vitamin B-12 deficiency in Giant Schnauzers.

Other nutritional influences include hip dysplasia in puppies fed excessive calories; osteochondritis dissecans (an inflammatory and degenerative bone and cartilage disorder) in dogs fed high levels of calcium; and hypercholesterolemia (high serum cholesterol) in inbred sled dogs fed high-fat diets.

Nutritional factors that play an important role in immune function include zinc, selenium and vitamin E, vitamin B-6 (pyridoxine) and linoleic acid. Deficiencies of these compounds impair both circulating (humoral) as well as cell-mediated immunity. The requirement for essential nutrients increases during periods of rapid growth or reproduction and also may increase in geriatric individuals, because immune function and the bioavailability of these nutrients generally wanes with aging.

As with any nutrient, however, excessive supplementation can lead to significant clinical problems, many of which are similar to the respective deficiency states of these ingredients. Supplementation with vitamins and minerals should only be given with the advice of a professional nutritionist and should not be viewed as a substitute for feeding premium-quality fresh and/or commercial dog foods.

BIBLIOGRAPHY

Dodds, W.J. "Autoimmune thyroid disease." DOG WORLD, 77 (4): pp. 36-40, 1992.

Dodds, W.J. "Unraveling the autoimmune mystery." DOG WORLD, 77 (5): pp. 44-48, 1992.

Dodds, W.J. "Genetically based immune disorders: Autoimmune diseases, Parts 1-3." *Veterinary Practice STAFF*, 4 (1, 2 and 3): pp. 8-10, 1, 26-31, 35-37, 1992.

Dodds, W.J. "Immune deficiency diseases: Genetically based immune disorders, Part 4." *Veterinary Practice STAFF*, 4 (5): pp. 19-21, 1992.

Dodds, W.J. "Vaccine safety and efficacy revisited." *Veterinary Forum*, May: pp. 68-71, 1993.

Berry, M.J., and P.R. Larsen. "The role of selenium in thyroid hormone action." *Endocrine Reviews*, 13 (2): pp. 207-219, 1992.

Ackerman, L. "The benefits of enzyme therapy." *Veterinary Forum*, October: pp. 45 and 69, 1993.

Tizard, I. *Veterinary Immunology: An Introduction*. 4th Ed. Philadelphia: W.B. Saunders Co., p. 498, 1992.

Dodds, W.J. and S. Donoghue. "Interactions of clinical nutrition with genetics." Chapter 8. *The Waltham Book of Clinical Nutrition of the Dog and Cat*. Oxford: Pergamon Press Ltd., 1994.

Cargill, J. and S. Thorpe-Vargas. "Feed that dog." Parts IV-VI. DOG WORLD, 78 (10-12): pp. 36-42, 28-31, 36-41, 1994.

W. Jean Dodds, D.V.M., is the president of HEMOPET, a national non-profit animal blood bank located in Irvine, Calif., and is a recognized authority in veterinary hematology and immunology.

Thyroid and Other Autoimmune Disorders of the Shih Tzu

by W. Jean Dodds, DVM

At the request of Barbara Pennington of the ASTC Committee for Immune-Mediated Diseases, the following commentary provides additional information in response to the recent timely article by Chris Jones (Shih Tzu Reporter, _____ 1990, pp. 93-94).

Early Thyroid Disease (Thyroiditis), Compensatory and Clinical Hypothyroidism

Most of the confusion about the diagnosis and treatment of thyroid disease in purebred or mixed breed dogs today stems from the expectation that affected animals must show clinical signs of inadequate thyroid hormonal production (i.e. hypothyroidism) in order to have this disease. The term hypothyroidism has been loosely applied to describe all stages of this disease process whereas strictly speaking it should be reserved for the end-stages when the animal's thyroid gland is no longer capable of producing sufficient hormone(s) to sustain clinical health. At this point, the dog can express any number of the non-specific multisystem signs of thyroid dysfunction. But lets start at the beginning --

The most common cause of canine thyroid disease is autoimmune thyroiditis (estimated 90% of cases). Thyroiditis is an immune-mediated process that develops in genetically susceptible individuals and is characterized by the presence of antithyroid antibodies in the blood or tissue. This condition usually, but not always, progresses eventually to thyroid disease (other commonly used terms for the human disease are "lymphocytic thyroiditis" or "Hashimoto's disease"). Thyroiditis is believed to start in most cases around puberty and gradually progresses through mid-life and old age to become clinically expressed hypothyroidism once thyroid glandular reserve has been depleted. During this process, the animal or person becomes more susceptible to immune-mediated or other diseases affecting various target tissues and organs [e.g., autoimmune hemolytic anemia (AIHA), idiopathic thrombocytopenic purpura (ITP), von Willebrand's disease (vWD), systemic lupus erythematosus (SLE); seizure disorders; chronic infections; immunosuppressive viral infections like distemper, parvovirus and retrovirus diseases; bone marrow failure; leukemia and lymphoma; chronic active hepatitis; immune eye, joint, kidney and adrenal (Addison's) diseases; chronic allergic and immune skin and muscle disorders]. The prerequisite genetic basis of this disorder has been proven in humans and more recently in dogs and several other species.

To better understand what happens in the transition from early thyroiditis to clinical hypothyroidism, a quotation from Dr. John E. Bethune, Chairman, Department of Medicine, UCLA is helpful (Disease-A-Month, 35, 1989). "It is important to clearly understand the changes that occur in thyroid physiology during the evolution of clinical hypothyroidism from the euthyroid state in humans. As primary thyroid gland failure develops and progresses, the free T4 concentration falls; in response to this, TSH is secreted from the pituitary gland, and its concentration in the serum rises. This stimulates the remaining thyroid tissue to make more T4 so that its level does not fall below the lower limit of normal. This also prevents the signs and symptoms of hypothyroidism from developing. This state of "compensated" hypothyroidism may last for some time and can be detected only by the raised TSH values. As the thyroid gland is stimulated, the conversion rate of T4 to T3 within the gland increases, and sufficient extra T3 is secreted to keep the level of free T3 normal for a considerable time, even after the free T4 level has fallen below the accepted normal range. As the thyroid mass decreases with progression of the disease state, the T4 concentration will finally fall below the normal range, and signs and symptoms of hypothyroidism will begin to appear. The T3 level remains normal until very late in the development of hypothyroidism."

The above explanation helps us to appreciate existing confusion and controversy within the veterinary profession regarding whether or not testing or treatment is indicated for dogs that fail to show so called "typical" signs of hypothyroidism. In fact, we have only recently begun to recognize the subtle signs of early thyroid dysfunction in dogs as the prevalence of the autoimmune form of the condition has increased within and among dog breeds. Today, some 50 breeds including the Shih Tzu are genetically predisposed to develop thyroid disease. Veterinary textbooks in print prior to 1990 and presumably even those recently printed are not up-to-date on this topic because the latest scientific and medical discoveries are only now reaching referreed publications and have therefore not appeared in textbooks.

Diagnosis of Thyroid Disease

1. Baseline Thyroid Profiles

A typical thyroid test measures the total presence of the T4 hormone in the body. But an important measurement is the small fraction of the total T4 which is not bound in the body and is therefore available to the tissues. This "free" T4 is converted in the tissues to T3 which is important for intracellular function. In the newer thyroid profiles available at specialized veterinary laboratories in the US and Canada, they test for total T4, total T3, free T4, cholesterol, and free T3.

Cholesterol should be measured from the same blood sample as the thyroid tests. In dogs with thyroid dysfunction the higher the cholesterol level, the lower the level of free T4. Owners can obtain a printout listing all the test results for their dog, as well as the "K" value, which is a linear constant between free T4 and cholesterol.

With respect to K values, those which are above +1 and preferably above +5 are considered to be normal. Less than -4 indicates an animal with primary thyroid disease, even if there are no overt symptoms. Values between -4 and +1 are either early thyroid disease and/or a nonthyroidal illness (another illness which depresses thyroid levels). Thyroid deficiency even at early subclinical stages can cause irreversible damage in the body, and the value of this more sensitive test is that problems can be detected before damage has become so severe that clear visible symptoms are present.

For a dog displaying symptoms of autoimmune disease, the K value can be helpful to indicate whether the dog has low, normal or high thyroid function, and if the autoimmune problems are likely to be responsive to thyroid hormone supplementation. If test results are normal, and the dog is nevertheless ill, proceed to do other diagnostic tests such as those for antithyroid antibodies [antithyroglobulin, anti T4 and anti T3 autoantibodies].

Michigan State University has recently introduced antithyroid hormone antibody (anti T4 and anti T3) testing as part of their complete thyroid panel. This is an important step towards more effective thyroid testing and results of nearly 16,000 samples screened to date indicate a substantially higher prevalence than previously reported. Contact the Endocrinology Section, Animal Health Diagnostic Laboratory, P.O. Box 30076, Lansing, MI 48909 (517-353-0621).

2. Thyroid Stimulating Hormone (TSH) Response Test

The thyroid profiling outlined above differs from the TSH response test often used to diagnose the clinical stages of hypothyroidism. This test measures thyroid reserve by assessing the ability of the thyroid gland to release T4 hormone upon stimulation with an injection of TSH. It is estimated that from 50-70% of thyroid follicular tissue has to be destroyed or damaged before hormonal reserves are depleted to the extent that overt hypothyroidism occurs. Thus, in the early stages of progressive thyroid disease (thyroiditis), the TSH response is usually normal and would be misleading (i.e., a false negative diagnosis). For this reason, we do not rely on finding a positive (abnormal) TSH response test to diagnose thyroiditis.

3. Antithyroglobulin Antibody (ATA) Test

Thyroglobulin is a glycoprotein found in thyroid gland follicles. The metabolites of thyroid hormones, T3 and T4, are incorporated in the primary structure of thyroglobulin. Thyroglobulin is antigenic and specific antibodies can form that are directed against several sites on the molecule. In humans, the presence of ATA is most often associated with Hashimoto's thyroiditis, the immune-mediated inflammation of the thyroid gland that progressively destroys functional thyroid tissue. The outcome of this process is hypothyroidism, and life-long treatment with thyroid hormone is required. Hashimoto's thyroiditis is common; and cases tend to cluster among families and certain ethnic groups. In humans, dogs, and other affected species, biopsy of the thyroid gland shows characteristic changes of lymphocytic infiltration and immunologic destruction.

Assays that measure ATA have been adapted for dogs. In two independent studies about 50-80% of the clinically hypothyroid dogs tested had significant levels of ATA. Positive ATA titers were also found in some dogs with nonthyroidal endocrine disease, and in some euthyroid relatives of hypothyroid dogs. At present, the prevalence of ATA in the various breeds affected with hypothyroidism, the value of ATA as a predictor of impending thyroid failure or a marker of familial hypothyroidism, and the relation of ATA to other immune-mediated disorders has not been determined. Preliminary studies of several affected dog families show promise in supporting the usefulness of ATA as predictive of subsequent hypothyroidism (e.g., Great Danes, Doberman Pinschers, Old English Sheepdogs and Shetland Sheepdogs). The quantitative ATA test if offered by the veterinary college at the University of Florida, Gainesville and as a research test by several groups including our own laboratory.

4. Genetic Screening for Thyroid Disease

These thyroid panels and antibody tests can also be used for genetic screening of apparently healthy animals to evaluate their fitness for breeding. A bitch with antithyroid antibodies in her blood may pass these along to her puppies in her colostral milk. Also, any dog having circulating antithyroid antibodies can eventually develop clinical symptoms of thyroid or other autoimmune diseases. Therefore, thyroid screening can be very important for potential breeding stock.

Thyroid testing for genetic screening purposes is less likely to be meaningful before puberty. Healthy young dogs (less than 15-18 months of age) should have thyroid baseline levels for all parameters in the upper third of the adult normal ranges. In fact, for optimum thyroid function in screening breeding stock, levels should be at least at the mid-point of the so-called

"normal" ranges, because lower levels may well be indicative of the early stages of thyroiditis among relatives of dog families known to have thyroid disease.

Treatment of Thyroid Disease

The new information summarized here has changed our approach to treatment and control of thyroid disease. In addition to providing thyroid supplementation for dogs showing the typical signs of thyroid disease, we now know that treatment of dogs showing the early stages of thyroiditis (based on the testing described above) is necessary and important to correct the underlying thyroid imbalance, reduce the risk of developing other related immune-mediated disorders (listed earlier), and to control or prevent the process of thyroiditis from progressing to depletion and exhaustion of the thyroid gland.

1. Type of Treatment

The treatment of choice because of its wide safety margin and efficacy is T4 hormone (L- or levothyroxine). The most commonly used brand names are Soloxine (Daniels) and Synthroid (Flint) and we recommend either of these over generics especially for the smaller breeds. Use of T3 hormone (triiodothyronine) is not recommended for initial use because toxicity can more easily develop with this product -- T3 is the intracellular hormone whereas most of T4 must be first converted to T3 before it achieves its metabolic effect. In some cases where the animal's body cannot properly convert T4 to T3, the dog will need both T4 and T3 therapy to correct the problem. For this purpose, the general rule of thumb is to give from 2/3 to a full dose of T4 and a 1/3 dosage of T3 [i.e., 0.1 mg per 10-20 pounds of T4 plus 1 ugm per pound of T3 twice daily]. However, no dog should be treated with these thyroid hormonal preparations without having proper veterinary testing, medical examination and followup. The article written by Chris Jones is excellent in emphasizing the importance of proper veterinary consultation.

2. Frequency of Treatment

Thyroid hormones should always be given twice daily to effect the best response. Until recently, veterinarians have been advised to give treatment to effect either once or twice daily because data on this point were unclear. We now know that the half-life of T4 in the dog is about 10-12 hours (much shorter than humans); for T3, it's only 6-8 hours. Thus, about half of the hormone is metabolized and excreted from the body within 12 hours. Furthermore, twice daily dosing aids in controlling thyroiditis because it shuts off pituitary production of TSH by negative feedback in concert with the half-life of the hormone. In other words, the dog's own thyroid follicular cells become quiescent and are less likely to stimulate production of the

antithyroid antibodies responsible for the disease. (Obviously these are simplistic explanations of the complex metabolic, immunologic and biochemical events involved). Contrary to some popular "wisdom", treatment with thyroid hormone does not destroy or suppress the potential of the gland to response on its own once treatment is stopped for whatever reason. The latest veterinary research shows that it takes the thyroid gland up to 30 days to recover its full hormonal potential once therapy is withdrawn. Therefore, if an animal has been medicated where the diagnosis is unclear, treatment should be withdrawn (if it's clinically safe to do so) for 30 days before the animal is retested with the complete type of thyroid profile described above.

Followup testing after initiating treatment is usually performed after 4-8 weeks of therapy. The sample should be taken 4-6 hours after the morning dosage and optimum results will show thyroid values in the upper third of normal ranges at that peak time of absorption. Dosage can then be adjusted accordingly if needed. Dogs on longterm therapy with thyroid hormones should be monitored with complete panels (not just T4 as you need to be sure the dog's body is converting the T4 medication properly to T3) on a regular basis (every 6-12 months).

Specific Comments Relevant to Shih Tzus

While the Shih Tzu may not have been listed as a specific breed predisposed to thyroid disease until recently, this reflects the lag time between generation of new information and publication dates of textbooks and journals (several years). The rapid increase in the number of commonly affected dog breeds worldwide attests to the recent change in both the recognition and prevalence of this disease. (A parallel increase has occurred in humans over the past decade). Veterinary experiences will naturally vary in different parts of the country, in urban versus rural settings and with the size of practice and number of Shih Tzu bred and seen within a particular area.

It is true that testing relatives of known affected families for a genetically transmitted or predisposed condition like thyroid disease will tend to bias the sample population in favor of finding more abnormal animals. However, reliable prevalence data within a breed can be estimated by collecting information from the major show bloodlines and pet-stock of a representative population across the country. Our experience to date with vWD and the autoimmune blood and thyroid diseases of Shih Tzus indicate increasing prevalence throughout the breed. These have become significant problems for the Shih Tzu. The primary underlying factor which predisposes the breed to these blood and other autoimmune disorders is autoimmune thyroid disease. A data base already exists but having more data is always helpful and provides an expanding perspective. We would

be happy to participate in collecting information from interested breeders; material sent is kept strictly confidential.

Additional Reading: Dodds, W.J. "1990 Update on Autoimmune Disorders of Purebred Dogs." Pure Bred Dogs American Kennel Gazette, Vol. 107 #7 (July), 86-94, 1990

W. Jean Dodds, DVM
President, HEMOPET
938 Stanford Street
Santa Monica, California 90403
(213) 828-4804

CANINE THYROID AND AUTOIMMUNE DISEASE

(Genetics, Nutrition and Vaccine-Related Issues)

W. Jean Dodds, DVM
Hemopet
938 Stanford Street
Santa Monica, CA 90403
310-828-4804; Fax 310-828-8251

Hypothyroidism is the most common endocrine disorder of canines. A recent survey of breed clubs conducted by the American Kennel Club Delegates Committee on health matters indicated that hypothyroidism was the most common health concern of the majority of purebred dog fanciers. An estimated 80% of the cases of canine hypothyroidism result from autoimmune (lymphocytic) thyroiditis. The heritable nature of this disorder poses significant genetic implications for breeding stock. Accurate diagnosis of the early compensatory stages of canine autoimmune thyroiditis leading up to hypothyroidism affords important genetic and clinical options for prompt intervention and case management.

Although thyroid dysfunction is the most frequently recognized endocrine disorder of pet animals, it is often difficult to make a definitive diagnosis. As the thyroid gland regulates metabolism of all body cellular functions, reduced thyroid function can produce a wide range of clinical manifestations (Table 1) Many of these clinical signs mimic those resulting from other causes and so recognition of the condition and interpretation of thyroid function tests can be problematic. Dr. David Panciera (1997) succinctly captured this situation in a recent editorial -- "a healthy dose of skepticism should accompany interpretation of any thyroid function test, with evaluation of the history and physical examination findings being paramount to an accurate diagnosis".

Tests Used for Diagnosing Thyroid Disease (Table 2)

Confusion remains over which diagnostic tests are the most specific and sensitive for identifying thyroid dysfunction especially in its early stages.The tests described below can also be used in other animal species, except where noted.

Older Tests for Thyroid Disease

1. Total T4

Measuring serum T4 (or T3) is not a reliable means for diagnosis of thyroid disease in any species, because it can : 1) overdiagnose hypothyroidism; 2) underdiagnose hyperthyroidism; and 3) fail to detect early stages of the compensatory disease and thyroiditis. This test is greatly influenced (lowered) by the presence of nonthyroidal illness (NTI) and specific drug therapy (e.g. corticosteroids, anticonvulsants, sulfonamides, nonsteroidal anti-inflammatory agents). For these reasons, measuring serum T4 or T3 alone or as the only thyroid analyte of a health profile is not recommended.

2. TSH Response Test

This dynamic test of thyroid function was considered the most reliable means of diagnosing clinical hypothyroidism, despite the fact that it only measures thyroid reserve and therefore fails to detect the early stages of thyroid disease. Today, the thyroid-stimulating hormone (TSH) response test is rarely performed because the bovine source of TSH used in the test is no longer marketed. An alternative dynamic test using TRH has recently been evaluated (see below).

Current and New Tests for Thyroid Disease

1. Free (Unbound) T4

Most veterinary diagnostic laboratories now offer comprehensive diagnostic tests for thyroid disease. The basic panel includes total T4, total T3, free T4, free T3, T3 autoantibody (T3AA) and T4 autoantibody (T4AA), which can be augmented with one or more of the newer tests described below. Accurate measurement of free T4 is the most important component of this analysis because it represents the biologically active (unbound) fraction of the total T4. Appropriate methods include the solid-phase analog and chemiluminescence assays and equilibrium dialysis (EQD), the latter being considered the "gold standard" against which the other two have been validated with a high degree of correlation (> 90%). By contrast, liquid-phase analog free T4 assays used routinely in human medicine are not reliable in animals (usually read too low). Even the EQD method of measuring free T4 can yield misleading results, because increases to very high levels can occur in nonthyroidal disease, and, if the serum gets too warm, bound T4 can dissassociate from its binding proteins thereby raising the unbound or free fraction.

On the plus side, free T4 assays are less likely to be influenced by NTI and drug therapy. Reliable (validated) methods for measuring this analyte are the most useful for accurate diagnosis of canine hypothyroidism, especially in the early stages or in the presence of NTI.

2. Endogenous Canine Thyroid Stimulating Hormone (cTSH)

Validated first generation assays for cTSH are now available and have also been used in cats. In primary hypothyroidism, as serum free T4 levels fall, pituitary output of TSH rises in a regulatory, compensatory response. Elevated serum TSH levels are therefore another indicator of thyroid dysfunction. Recent experience with these tests, however, indicates about 20-38% discordancy between expected and actual results in normal dogs as well as confirmed cases of hypothyroidism or NTI. Furthermore, a recent study showed that it takes 40 separate measurements to accurately determine the basal TSH levels of healthy dogs. With respect to NTI, humans with chronic renal disease (CRD) have a much higher incidence of true hypothyroidism than age-matched control people, although TSH levels using third generation tests are also increased in other CRD patients that are euthyroid. Until these first generation TSH assays for animals are refined to improve their predictive capacity, they should not be relied upon as the sole or major basis for interpreting thyroid function status.

3. Canine Thyroglobulin Autoantibodies (TgAA)

An estimated 80% of cases of canine hypothyroidism result from heritable autoimmune (lymphocytic) thyroiditis. In most of these cases, TgAA are present in the serum, whereas only about 20% of cases of thyroiditis have elevated circulating T3 and/or T4 AA. Thus, the presence of elevated T3 and/or T4 AA confirms a diagnosis of autoimmune thyroiditis but underestimates its prevalence, as negative (non-elevated) autoantibody levels do not rule out thyroiditis. Measuring TgAA levels also permits early recognition of the disorder, and facilitates genetic counselling as affected dogs should not be bred. A commercial TgAA test is available and has been validated with a representative number of field cases from the various breeds affected with autoimmune thyroid disease. It can give false negative results if the dog has received thyroid supplement within the previous 90 days, thereby allowing unscrupulous owners to test dogs while on treatment to obtain certification with health registries such as the Orthopedic Foundation of America (OFA)Thyroid Registry. [To be certified, the OFA requires normal values for free T4 by EQD, cTSH, and TgAA, with retesting every two years.] Furthermore, false positive results may be obtained if the dog has been vaccinated within the previous 30-45 days, or in some cases of NTI.

4. TRH Response Test

A new dynamic test of thyroid function, the thyrotropin-releasing hormone (TRH) response test has been promoted as a useful replacement for the TSH response test. However, inconsistent increases in T4 have been noted following TRH stimulation in normal dogs, and this test failed to distinguish hypothyroid from euthyroid dogs with dermatopathies. On the otherhand, measuring the serum endogenous TSH level after TRH administration has recently been shown to help evaluate thyrotroph function. It may be particularly useful in dogs with concurrent clinical signs of pituitary or hypothalamic disease (e.g. central vision impairment, circling, head pressing) to confirm secondary or tertiary hypothyroidism.

5. Baseline Thyroid Profiles

The normal reference ranges for thyroid analytes of healthy adult animals tend to be similar for most breeds of companion animals. Exceptions are the sighthound and giant breeds of dogs which have lower basal levels. Typical thyroid levels for healthy sighthounds, such as retired racing greyhounds, are at or just below the established laboratory reference ranges, whereas healthy giant breeds have optimal levels around the midpoint of these ranges.

Similarly, because young animals are still growing and adolescents are maturing, optimal thyroid levels are expected to be in the upper half of the references ranges. For geriatric animals, basal metabolism is usually slowing down, and so optimal thyroid levels are likely to be closer to midrange or even slightly lower. As stated above, a complete baseline thyroid profile typically has total T4, total T3, free T4, free T3, T3AA and T4AA, and also can include cTSH and/or TgAA. The TgAA assay is especially important in screening breeding stock for heritable autoimmune thyroid disease (see below).

Genetic Screening for Thyroid Disease

The majority of canine thyroid disease is due to autoimmune thyroiditis, which is a familial disorder of inherited predisposition similar to that of human Hashimoto's disease. Therefore, the baseline thyroid profile used for thyroid testing must include assays for thyroid AA (T4AA, T3AA, and/or TgAA). This profile can be applied not only to clinical patients suspected of having thyroid disease, but also can be used for genetic screening of apparently healthy relatives to evaluate their fitness for breeding. A bitch with circulating thyroid AA has the potential to pass these along to the

puppies transplacentally as well as via the colostrum. Furthermore, any dog having thyroid AA may eventually develop clinical symptoms of thyroid disease and/or be susceptible to other autoimmune diseases. Thyroid screening is thus very important for selecting potential breeding stock as well as for clinical diagnosis.

Thyroid testing for genetic screening purposes is less likely to be meaningful before puberty. Screening is initiated, therefore, once healthy dogs and bitches have reached sexual maturity (between10-14 months in males and during the first anestrous period for females following their maiden heat). As the female sexual cycle is quiescent during anestrus, any influence of sex hormones on baseline thyroid function will be minimized. This period generally begins 12 weeks from the onset of the previous heat and lasts one month or longer. The interpretation of results from baseline thyroid profiles in intact females will be more reliable when they are tested in anestrus. In fact, genetic screening of intact females for other disorders such as von Willebrand disease (vWD), hip dysplasia, and wellness or reproductive checkups (vaginal cultures, hormone testing) is best scheduled during anestrus. Once the initial thyroid profile is obtained, dogs and bitches should be rechecked on an annual basis to assess their thyroid function and overall health. Generation of annual test results provides comparisons that permit early recognition of developing thyroid dysfunction. This allows for early treatment, where indicated, to avoid the appearance or advancement of clinical signs associated with hypothyroidism.

For optimal health, puppies and adolescent dogs under 15-18 months of age should have baseline thyroid levels in the middle to upper half of adult normal ranges. This pertains to young dogs because they are still growing and maturing, and so require higher levels of thyroid hormones to complete development. Similarly, in older animals above 8-10 years of age, body functions start to slow down and so baseline thyroid levels may be in the lower half of the range in the euthyroid individual. For healthy young adults used for performance or breeding, optimum thyroid function should be at least at the mid-point of the established normal ranges for the particular laboratory used. Lower levels, even in the absence of clinical signs compatible with thyroid disease, may be indicative of the early stages of thyroiditis, especially if they are found in relatives of dogs from families previously documented to have thyroid disease.

Diagnosing Difficult or Equivocal Cases

The difficulty in accurately diagnosing some cases of thyroid disease is compounded by the fact that certain patients with typical clinical signs of hypothyroidism have circulating levels of thyroid hormones within the normal range. Many will improve clinically when given thyroid medication, because blood levels of thyroid hormones may not reflect their cellular and tissue levels. A 6-8 week clinical trial of thyroid supplement given BID is safe and appropriate for such patients, and is followed by rechecking the complete thyroid profile 4-6 hours after the morning pill. Response to thyroid therapy is considered an appropriate justification to continue thyroid therapy along with annual checkups to adjust dosages as appropriate.

If an animal is receiving thyroid supplement and the basis for the original diagnosis is unclear, the clinician may elect to discontinue therapy and retest. In such cases, or when thyroid therapy is discontinued for any other reason, retesting is performed after another 6 weeks. (It takes this long for the pituitary-thyroid axis to be restored to full productive capacity after cessation of thyroid therapy.) To perform thyroid retesting before this period has elapsed would yield relatively low blood levels of thyroid hormones thereby biasing the interpretation in favor of hypothyroidism. Once the appropriate dose of thyroid supplement is established, annual retesting is recommended.

Canine Autoimmune Thyroiditis

In recent years the prevalence of autoimmune (immune-mediated) diseases has been increasing rapidly in humans and animals. Scientists and clinicians have attributed this increase to such factors as genetic and sex predisposition, nutritional influences, exposure to toxins and drugs, recent viral infections or use of polyvalent vaccines, and pituitary-thyroid axis imbalances. Today, an estimated 80% of cases of canine hypothyroidism result from autoimmune (lymphocytic) thyroiditis. The heritable basis of this disorder poses significant genetic implications for breeding stock, and affected animals should not be used for breeding.

This author has compiled and analyzed 1060 canine cases of autoimmune thyroiditis between January 1995 and January 1999 (Table 3). Purebreds made up 96% of this group, and both sexes were equally represented. The mean age at diagnosis was 4.4 years (range 2.6-12.0), and mean levels of T3AA and T4AA were 6.0 (range 1.2-22.2) and 1.6 (range 0.6-8.8), respectively, with normal reference levels being below 2. The most prevalent circulating thyroid AA was against T3

(996/1060 cases; 94%) and 254/1060 (24%) had combined T3 and T4AA. In a few instances (51/723;7%), the dogs demonstrated only T4 autoantibodies. The twelve breeds most affected were the Golden Retriever (209 cases), Shetland Sheepdog (124), American Cocker Spaniel (68), Boxer (51), Doberman Pinscher (42), Labrador Retriever (40), German Shepherd (19), Akita (15), Irish Setter (14), English Setter (13), Old English Sheepdog (12), and Collie (10), although many other breeds and mixed breeds were also represented (Table 3). All of these dogs also would be expected to have elevated levels of TgAA. In fact, all 75 dogs selected for testing at random from this case cohort had high TgAA levels.

Polyglandular Autoimmunity

Individuals genetically susceptible to autoimmune thyroid disease may also become more susceptible to immune-mediated diseases affecting other target tissues and organs, especially the bone marrow, liver, adrenal gland, pancreas, skin, kidney, joints, bowel, and central nervous system. The resulting "polyglandular autoimmune syndrome" of humans is becoming more commonly recognized in the dog, and probably occurs in other species as well. The syndrome tends to run in families and is believed to have an inherited basis. Multiple endocrine glands and nonendocrine systems become involved in a systemic immune-mediated process. This multiple endocrinopathy often occurs in patients with underlying autoimmune thyroid disease (hypo- or hyperthyroidism) and concurrent Addison's disease, diabetes, reproductive gonadal failure, skin disease and alopecia, and malabsorption syndrome. The most common nonendocrinologic autoimmune disorders associated with this syndrome are autoimmune hemolytic anemia (AIHA), idiopathic thrombocytopenic purpura (ITP), chronic active hepatitis, and immune-complex glomerulonephritis (systemic lupus erythematosus; SLE).

The most commonly recognized polyglandular endocrinopathy of dogs is Schmidt's syndrome (thyroiditis and Addison's disease). Dog breeds genetically predisposed to this disorder include the Standard Poodle, Old English Sheepdog, Bearded Collie, Portuguese Water Dog, Nova Scotia Duck Tolling Retriever, and Leonberger, although any breed or mixed breed can be affected. Our study cohort of 162 cases of autoimmune blood and endocrine disorders in Old English Sheepdogs (1980-1989) included 115 AIHA and/or ITP, 99 thyroid disease, 23 Addison's disease, 7 vaccine reactions, 3 SLE, 2 diabetes, 1 rheumatoid arthritis and 1 hypoparathyroidism. The group

comprised 110 females (15 spayed) and 52 males (3 neutered). Seven of the most recent 103 cases had two or more endocrine disorders, and 101 of the 108 cases where pedigrees were available showed a familial relationship going back several generations. Data from surveying the Bearded Collie breed reported 55 hypothyroid, 17 Addison's disease, and 31 polyglandular autoimmunity (5 were hypothyroid).

Aberrant Behavior and Thyroid Dysfunction

An additional noteworthy clinical finding in dogs affected with thyroid or polyglandular autoimmune disease has been the sudden or progressive onset of aberrant behavior including aggression, submissiveness, shyness, fearfulness, passivity, seizure disorder, excitability, sensitivity to noise, anxiety, irritability, compulsiveness, chewing, moodiness, lethargy, depression, and unstable temperament. A similar association between behavioral and psychologic changes and thyroid dysfunction has been recognized in humans since the 19th century, and more recently has been noticed in cats with hyperthyroidism. In a recent human study, 66% of patients with attention deficit- hyperactivity disorder were found to be hypothyroid, and supplementing their thyroid levels was largely curative.

Tables 4-5 summarize results of complete thyroid diagnostic profiling on 634 canine cases of aberrant behavior, compiled by this author in collaboration with Drs. Nicholas Dodman, Linda Aronson, and Jean DeNapoli of Tufts University School of Veterinary Medicine, North Grafton, MA. Ninety percent (568 dogs) were purebreds and 10% were mixed breeds. There was no sex predilection found in this case cohort, whether or not the animals were intact or neutered. Sixty-three percent of the dogs had thyroid dysfunction as judged by finding 3 or more abnormal results on the comprehensive thyroid profile. The major categories of aberrant behavior were aggression (40% of cases), seizures (30%), fearfulness (9%), and hyperactivity (7%); some dogs exhibited more than one of these behaviors (Table 5). Within these 4 categories, thyroid dysfunction was found in 62% of the aggressive dogs, 77% of seizuring dogs, 47% of fearful dogs, and 31% of hyperactive dogs.

Outcomes of treatment intervention with standard twice daily doses of thyroid replacement were evaluated in 95 cases. Of these, 58 dogs had greater than 50% improvement in their behavior as judged by a predefined 6-point subjective scale (34 were improved > 75%), and another 23 dogs had >25 but <50% improvement. Only 10 dogs experienced no appreciable change, and 2 dogs had

a worsening of their behavior. When compared to 20 cases of dominance aggression treated with conventional behavior or other habit modification over the same time period, only 11 dogs improved more than 25%, and of the remaining 9 cases, 3 failed to improve and 3 were euthanized or placed in another home. These initial results are so promising that complete thyroid diagnostic profiling and treatment with thyroid supplement, where indicated, is warranted for all cases presenting with aberrant behavior.

Von Willebrand Disease and Thyroid Metabolism

An acquired form of vWD associated with hypothyroidism has been recognized in humans and several dogs breeds, particularly the Doberman Pinscher, but also the Shetland Sheepdog, Rottweiler and Golden Retriever. The relationship is complex and has yet to be fully elucidated at the molecular level. Clearly, not all humans or dogs with hypothyroidism have low levels of von Willebrand factor (vWF), and not all those with low vWF have hypothyroidism. Asymptomatic vWD carriers often express a bleeding tendency in mid-life when they become hypothyroid. Development of hypothyroidism apparently exacerbates their original trait and compromises hemostasis. In our own studies, the average age of onset of bleeding signs in Dobermans Pinchers was over 4 years. The question appears to relate to the metabolic regulation of vWF biosynthesis and secretion rather than plasma levels or even platelet adhesiveness as measured by the mucosal bleeding time.

In other breeds there are insufficient data at present to causally link vWD with hypothyroidism, despite the fact that most (45 or more) of the 59 or more breeds recognized to have vWD also suffer from familial hypothyroidism. Therefore, the causal relationship established for several breeds does not necessarily apply to all affected breeds. In the Irish Wolfhound, more than half of the several hundred dogs screened to date at their annual National Specialty shows have had low vWF levels and about one quarter have had low thyroid parameters. However, when these data were analyzed there was a poor correlation between the two findings. Perhaps only those dogs that develop a particular type of thyroid disease (e.g. autoimmune thyroiditis with elevated TgAA or T3/T4 AA) are the ones more likely to express an acquired form of vWD as adults.

The more definitive way to study this relationship has been in thyroidectomized individuals, and those with congenital hypothyroidism or clinically expressed vWD and hypothyroidism. Results

of these studies support the fact that hemostatic function is altered when thyroid metabolism is impaired, although the mechanism is not understood. This defect in hemostatic function is rapidly corrected (within 12-24 hours) by giving standard doses of thyroid supplement.Regardless, the debate about the nature and extent of this physiologic and pathologic relationship is likely to continue because of its underlying clinical significance.

Other Factors Influencing Thyroid Metabolism

1. Nutritional Factors

Nutritional influences can have a profound effect on thyroid metabolism. The classical example is the iodine deficiency that occurs in individuals eating cereal grain crops grown on iodine-deficient soil. This will impair thyroid metabolism because iodine is essential for formation of thyroid hormones. Iron and zinc also are important minerals in regulating thyroid metabolism. Another important link has recently been shown between selenium deficiency and hypothyroidism. Cereal grain crops grown on selenium-deficient soil will contain relatively low levels of selenium. While commercial pet food manufacturers compensate for variations in basal ingredients by adding vitamin and mineral supplements, it is difficult to determine optimum levels for so many different breeds of animals having varying genetic backgrounds and metabolic needs.

The selenium-thyroid connection has significant clinical relevance, because blood but not tissue levels of thyroid hormones rise in selenium deficiency. Thus, selenium-deficient individuals showing clinical signs of hypothyroidism could be overlooked on the basis that blood levels of thyroid hormones appear normal. The selenium issue is further complicated because the synthetic antioxidants used in foods to protect fats from rancidity can impair the bioavailability of vitamin A, vitamin E and selenium, and alter cellular membrane function, metabolism and detoxification.

Because animals with autoimmune thyroid disease have generalized metabolic imbalance and often have associated immunological dysfunction, it is advisable to minimize their exposures to unnecessary drugs, chemicals and toxins, and to optimize their nutritional status with healthy balanced diets. Families of dogs susceptible to thyroid and other autoimmune diseases show generalized improvement in health when fed premium cereal-based diets preserved naturally with vitamins E and C rather than with the synthetic chemical antioxidants such as ethoxyquin, BHA and BHT. Fresh vegetables cooked with Italian herbs and garlic, dairy products such as yogurt or low

fat cottage cheese, or meats such as lamb, rabbit, venison, chicken and turkey can be added as supplements.

2. Infectious Agents and Vaccines (also see below)

Challenging the immune system of animals affected by thyroid disorders with infectious diseases or polyvalent modified-live vaccines has been associated with adverse effects in some cases. General recommendations are: to reduce exposures to contagious diseases and allergens; avoid booster vaccinations of geriatric patients, and during times of illness, recovery or relapses; use killed vaccine products, when these are available, and space vaccines at least 10-14 days apart to avoid excessive antigenic challenge; or perform serum antibody titration (vaccine titers) as an alternative to booster vaccination to assess the adequacy of existing immune memory. In some affected dog families, use of polyvalent combination modified-live vaccines has apparently induced seizure disorders, AIHA and/or ITP, bone marrow failure, acute fevers, and renal failure leading to amyloidosis, lameness, stiffness and arthritic pain to the extent that many of these dogs cannot stand up or move. This is just one example of the general principal to avoid unduly challenging susceptible animals during periods of rapid growth, hormonal change, or stress events.

3. Drugs

Many drugs are known to affect thyroid function. They produce their effects by various mechanisms including: 1) decreasing TSH secretion (e.g. steroids, dopamine); 2) decreasing thyroid hormone secretion (e.g. sulfonamides, lithium, iodide, amidarone); 3) decreasing T4 absorption (e.g. sucralfate, ferrous sulfate, aluminum hydroxide); 4) decreasing T3 and T4 transport in serum (e.g. estrogens, mitotane, androgens, steroids, furosemide, salicylates); and 5) increasing T3 and T4 metabolism (e.g. phenobarbital, rifampin, phenytoin, steroids, amidarone).

4. Chemical (Xenobiotic) Exposures

The ability of the body to handle chemical exposures to such compounds as polybrominated biphenyls, phenolics including bioflavonoids, chlorinated compounds, goitrogens, and the detoxification of drugs and chemicals via cytochrome P_{450} pathways all depend upon adequate and sustained thyroid function.

Thyroid Therapy

The mean residence time of thyroid hormone in companion animal species varies from 12-16 hours. The rationale for supplementing hypothyroid animals twice, rather than once daily, is partly

based on this fact but also is supported by the improved clinical response observed when the daily metabolic requirement is split into two equal doses. Most clinicians use synthetic L-thyroxine, preferably of brand name rather than generic source. The standard dose is 0.1 mg per 10 lbs or 4.5 kg BID for dogs, other than giant breeds or sighthounds, in which case the dosage is reduced to 0.1 mg per 15-20 lbs or 6.5-9 kg BID. For geriatric dogs, the lower dosage is recommended because of their less active metabolism. Generally, even giant breed dogs are started at no more than 0.8 mg BID and then increased from there, if needed. Post-pill thyroid monitoring should be performed after a 6-8 week therapeutic trial, taking the sample 4-6 hours after the morning dosage, and then repeated annually. Typically, the need for thyroid replacement is lifelong, although an occasional human or canine case can go into complete remission.

Holistic practitioners may opt to treat thyroid disorders with natural glandular products and/or to boost thyroid function with nutritional and herbal support. In this author's experience, thyroid or pituitary-thyroid combination glandulars have given uneven clinical responses, especially in larger dogs. For this reason, we prefer to use synthetic L-thyroxine BID to regulate thyroid function, and then use complementary approaches to control related or concurrent clinical problems. These include minimizing and detoxifying environmental exposures; balancing dietary trace mineral, vitamin and fatty acid needs; and feeding hypoallergenic diets along with digestive support.

Treating Refractory Cases or Circulating Thyroid Autoantibodies

1. Use of T3 Supplement

When clinical signs of thyroid disease are only partially or poorly ameliorated by supplementation with L-thyroxine at the standard dosages listed above, combination therapy is often successful. In these cases, the T4 supplement may be poorly converted to T3 by the liver and other tissues, so that addition of a T3 supplement or a thyroid glandular product containing both T3 and T4 is indicated. The typical treatment regimen includes the full dosage of T4 supplement given BID plus 1 μgm per lb or 2.2 μgm per kg of T3 supplement (Cytomel®) given BID or TID. This combination has been particularly beneficial for patients with concomitant liver disease or dysfunction, because the liver is the primary site of conversion of T4 to T3 and so conversion may be impaired in the presence of the hepatocellular disease.

The other situation applies to patients on anticonvulsant therapy for seizure disorders. Providing a low dosage of T3 supplement helps maintain adequate levels of T3 in the central nervous

system and may assist in raising the seizure threshold. Experience with this approach indicates that the addition of T3 supplement may allow the dosage of anticonvulsant required for seizure control to be lowered or even discontinued. A second advantage could be to offset any adverse effects of anticonvulsants on liver metabolism which could impair hepatocellular conversion of T4 to T3.

2. Reversal of Thyroid Autoantibodies

For patients with circulating T4 and/or T3 AA, even in the absence of typical clinical signs of thyroid disease, the rationale for thyroid supplementation is to interrupt the progression of thyroiditis and reverse the stimulus for production of thyroid AA. Experience with over 100 cases followed periodically for up to 6 years indicates that it takes between 5-7 months of thyroid replacement to cause levels of circulating thyroid AA to wane progressively and then disappear. Occasional cases never completely reverse AA production. We do not recommend using these animals in a breeding program, because of the heritable nature of thyroiditis.

Supplementation with L-thyroxine is believed to reverse the production of circulating thyroid AA by either inducing immune tolerance and/or by negative feedback inhibition of TSH and its effects on the TSH receptor. In a typical case, the standard therapeutic dose of L-thyroxine is given BID for 8-12 weeks. At that point the complete baseline thyroid profile is measured again to determine whether thyroid AA levels are waning. Patients rarely need retesting prior to this time because the presence of thyroid AA interferes with the accurate measurements of T3 and/or T4, and so little is gained from the additional cost to the client of rechecking at an earlier time point. Other clinical problems, such as pruritic skin disease, are treated with conventional or alternative approaches.

Vaccine-Related Issues

Changing Vaccine Protocols

The challenge to produce effective and safe vaccines for the prevalent infectious diseases of humans and animals has become increasingly difficult. In veterinary medicine, evidence implicating vaccines in triggering immune-mediated and other chronic disorders (vaccinosis) is compelling. While some of these problems have been traced to contaminated or poorly attenuated batches of vaccine that revert to virulence, others apparently reflect the host's genetic predisposition to react adversely upon receiving the monovalent or polyvalent products given routinely to animals. Animals

of certain susceptible breeds or families appear to be at increased risk for severe and lingering vaccine reactions. Recent studies also implicate vaccines in triggering autoimmune thyroiditis.

The onset of adverse reactions to conventional vaccinations (or other inciting drugs, chemicals, or infectious agents) can be an immediate hypersensitivity or anaphylactic reaction, or can occur acutely (24-48 hours afterwards), or later on (10-30 days) in a delayed type immune response usually caused by immune-complex formation. Typical signs of adverse immune reactions include fever, stiffness, sore joints and abdominal tenderness, susceptibility to infections, central and peripheral nervous system disorders or inflammation, collapse with autoagglutinated red blood cells and jaundice, or generalized pinpoint hemorrhages or bruises. Liver enzymes may be markedly elevated, and liver or kidney failure may accompany bone marrow suppression. Furthermore, recent vaccination of genetically susceptible breeds has been associated with transient seizures in puppies and adult dogs, as well as a variety of autoimmune diseases including those affecting the blood, endocrine organs, joints, skin and mucosa, eyes, muscles, liver, kidneys, and bowel. The underlying genetic basis of these conditions places other littermates and close relatives at increased risk.

Vaccination also can overwhelm the immunocompromised or even healthy host that is repeatedly bombarded with other environmental stimuli and is genetically predisposed to react adversely upon viral challenge. The recently weaned young puppy or kitten entering a new environment is at greater risk here, as its immature immune system can be temporarily or more permanently harmed. Consequences in later life may be the increased susceptibility to chronic debilitating diseases.

As combination (polyvalent) vaccines contain antigens other than the clinically important infectious disease agents, some may be unnecessary, and their use may increase the risk of adverse reactions. Today's licensed leptospirosis bacterins afford little, if any, protection against the clinically important fields strains, and the antibodies they elicit last only a few months. Other vaccines, such as for Lyme disease, may not be needed, because the disease is limited to certain geographical areas. Annual revaccination for rabies is required by some states even though USDA licensed rabies vaccine have a 3-year duration. Thus, the overall risk-benefit ratio of using certain vaccines or multiple antigen vaccines given simultaneously and repeatedly should be reexamined. It must be recognized, however, that the luxury of asking such questions today is presented only because the risk of disease has been effectively reduced by the widespread use of vaccination programs.

Given this troublesome situation, what are the experts saying about these issues? In 1995, a landmark review commentary focused the attention of the veterinary profession on the advisability of current vaccine practices. Are we overvaccinating companion animals, and if so, what is the appropriate periodicity of booster vaccines ? Discussion of this provocative topic generally leads to other questions about the duration of immunity conferred by the currently licensed vaccine components.

In response to questions posed above, veterinary vaccinologists have recommended new protocols for dogs and cats. These include: 1) giving the puppy or kitten vaccine series followed by a booster at one year of age; 2) administering further boosters in a combination vaccine every three years or as split components alternating every other year until; 3) the pet reaches geriatric age, at which time booster vaccination is likely to be unnecessary and may be unadvisable for those with aging or immunologic disorders. In the intervening years between booster vaccinations, and in the case of geriatric pets, circulating humoral immunity can be evaluated by measuring serum vaccine antibody titers as an indication of the presence of "immune memory". This latter phrase is more correct than "protective immunity", because protection against disease means survival after challenge with the infectious agent and may not correlate with the serum antibody titer. Titers do not distinguish between immunity generated by vaccination or exposure to the disease, although the magnitude of immunity produced by vaccination is usually lower. Except where vaccination is required by law, animals that previously experienced an adverse reaction to vaccination or are at genetic or physiological risk for such reactions also can have serum antibody titers measured annually instead of revaccination. If adequate titers are found, the animal should not need revaccination until some future date. Rechecking antibody titers can be performed thereafter, or can be offered as an alternative to pet owners who prefer not to follow the conventional practice of annual or semiannual vaccination. Reliable serologic vaccine titering is available from several university and commercial laboratories and the cost is reasonable.

Relatively little has been published about the duration of immunity following vaccination, although new data are beginning to appear. In Sweden, an in-depth study found adequate titers against canine distemper virus (CDV) in 83% of a very large group of dogs vaccinated more than 4 years beforehand. Another recent study of dogs vaccinated 9-55.5 months previously found 73% of 122 dogs to have protective canine parvovirus (CPV) titers, and 79% of 117 dogs to be adequately protected against CDV. The authors concluded that annual revaccination should be maintained,

because less than 90% of those vaccinated reached their criteria for protective titers. However, using similar criteria to assess vaccine antibody titers in a larger group of dogs, we came to a different conclusion (Twark and Dodds, in press, 1999). Our study evaluated 1441 dogs for CPV antibody titer and 1379 dogs for CDV antibody titer. Of these, 95.1 % were judged to have adequate CPV titers, and nearly all (97.6 %) had adequate CDV titers. Vaccine histories were available for 444 dogs (CPV) and 433 dogs (CDV) (Table 7). Only 43 dogs had been vaccinated within the previous year, with the majority of dogs (268 or 60%) having received a booster vaccination 1-2 years beforehand. On the basis of our data, we concluded that annual revaccination is unnecessary.

The vaccine histories obtained for this study indicate that the majority of serum samples were submitted at a time when annual booster vaccines typically would be given. When correlated with the high incidence of adequate immune memory found in this population of dogs, this study supports the belief that annual vaccination for CDV and CPV may not be necessary to provide adequate immune memory in the face of exposure to a particular virus. When an adequate immune memory has already been established, there is little reason to introduce unnecessary antigen, adjuvant, and preservatives by administering booster vaccines. By titering annually, one can assess whether a given animal's humoral immune response has fallen below levels of adequate immune memory. In that event, an appropriate vaccine booster can be administered.

A multifaceted approach to furthering the recognition of this situation, along with alternative strategies for containing infectious diseases and reducing the environmental impact of conventional vaccines is clearly needed. As a beginning we can increase the interval between adult booster vaccinations from one to three years, except as required by law, and monitor serum antibody levels for assessing immune memory response to the clinically important infectious agents.

References

Antech Diagnostics News. Changing Vaccine Protocols. April 1998. Ibid. Canine Thyroid Disease. November 1998.

Avgeris S, Lothrop CD Jr, McDonald TP. Plasma von Willebrand factor concentration and thyroid function in dogs. J Am Vet Med Assoc 196: 921-924, 1990.

Brooks M, Dodds WJ, Raymond SL. Epidemiologic features of von Willebrand's disease in Doberman Pinschers, Scottish Terriers, and Shetland Sheepdogs: 260 cases (1984-1988). J Am Vet Med Assoc 200: 1123-1127, 1992.

Bunevicius R, Kazanavicius G, Zalinkevicius R, et al. Effects of thyroxine as compared with thyroxine plus triiodothyronine in patients with hypothyroidism. N Eng J Med 340:424-429, 1999.

Burr H, Coyne M, Gay, C, et al. Duration of Immunity in Companion Animals after Natural Infection and Vaccination. Pfizer Animal Health 1998; pp 1-21.

Carmichael LE. Vaccines for dogs. In: Pastoret PP, Blancou J, Vannier P, et al, eds. Veterinary Vaccinology. Amsterdam: Elsevier, 1997; pp 326-335.

Carmichael LE. Canine viral vaccines at a turning point - a personal perspective. Adv Vet Med 41:289-307, 1999.

Chastain CB. Infectious diseases:canine leptospirosis. Ed Annot. Adv Sm Anim Med Surg 10(6):5, 1997.

Dewey CW, Shelton GD, Bailey, CS. Neuromuscular dysfunction in five dogs with acquired myasthenia gravis and presumptive hypothyroidism. Prog Vet Neurol 6: 117-123, 1995.

Dixon RM, Graham PA, Mooney CT. Serum thyrotropin concentrations: a new diagnostic test for canine hypothyroidism. Vet Rec 138: 594-595, 1996.

Dodds WJ. Genetically based immune disorders: Autoimmune diseases, Parts 1-3. Vet Pract STAFF 4 (1, 2, and 3): 8-10, 1, 26-31, 35-37, 1992.

Dodds WJ, Raymond SL, Brooks MD. Inherited and acquired von Willebrand's disease. Vet Pract STAFF 5(4) and 5(5): 1, 14-17; 21-23, 1993.

Dodds WJ. Hypothyroidism and von Willebrand factor. J Am Vet Med Assoc 206: 594-595, 1995.

Dodds WJ. Estimating disease prevalence with health surveys and genetic screening. Adv Vet Sci Comp Med 39: 29-96, 1995.

Dodds WJ. Autoimmune thyroiditis and polyglandular autoimmunity of purebred dogs. Can Pract 22 (1): 18-19, 1997.

Dodds WJ. What's new in thyroid disease ? Proc Am Hol Vet Med Assoc 1997; pp 82-95.

Dodds WJ. Holistic approaches for immune-mediated disease. Proc Am Hol Vet Med Assoc 1997; pp 96-100.

Dodds WJ. More bumps on the vaccine road. Adv Vet Med 41:715-732, 1999.

Duval D, Giger U. Vaccine-associated immune-mediated hemolytic anemia in the dog. J Vet Int Med 10:290-295, 1996.

Feldman EC, Nelson RW. Canine and Feline Endocrinology and Reproduction. Second Edition. Philadelphia: WB Saunders Co, 1996.

Frank LA. Comparison of thyrotropin-releasing hormone (TRH) to thyrotropin (TSH) stimulation for evaluating thyroid function in dogs. J Am An Hosp Assoc 32: 481- 487, 1996.

Hall IA, Campbell KC, Chambers MD, et al. Effect of trimethoprim-sulfamethoxazole on thyroid function in dogs with pyoderma. J Am Vet Med Assoc 202:1959-1962, 1993.

Happ GM. Thyroiditis - A model canine autoimmune disease. Adv Vet Sci Comp Med 39: 97-139, 1995.

Hogenesch H, Azcona-Olivera J, Scott-Moncreiff C, et al. Vaccine-induced autoimmunity in the dog. Adv Vet Med 41: 733-744, 1999.

Hoskins JD, Gourley KR, Taylor HW, et al: Challenge trial of Progard-7 vaccine using three field strains of CPV-2 Intervet's Companion Animal Technical Report 4:1- 4, 1995.

Hustead DR, Carpenter T, Sawyer DC, et al. Vaccination issues of concern to practitioners. J Am Vet Med Assoc 214: 1000-1002, 1999.

International Symposium on Canine Hypothyroidism, University of California, Davis. Can Pract 22 (1) : 4-62, 1997.

Iverson L, Jensen AL, Høier R, et al. Biological variation of canine serum thyrotropin (TSH) concentration. Vet Clin Pathol 28:16-19, 1999.

Jensen AL, Iversen L, Høier R, et al. Evaluation of an immunoradiometric assay for thyrotropin in serum and plasma samples of dogs with primary hypothyroidism. J Comp Pathol 114: 339-346, 1996.

Kaptein EM. Thyroid hormone metabolism and thyroid diseases in chronic renal failure. End Rev 17:45-63, 1996.

Mansfield PD. Vaccination of dogs and cats in veterinary teaching hospitals in North America. J Am Vet Med Assoc 208:1242-1247, 1996.

McGaw DL, Thompson M, Tate, D, et al. Serum distemper virus and parvovirus antibody titers among dogs brought to a veterinary hospital for revaccination. J Am Vet Med Assoc 213: 72-75, 1998.

McGregor AM. Autoimmunity in the thyroid - Can the molecular revolution contribute to our understanding? Quart J Med 82 (297): 1-13, 1992.

Nachreiner RF, Refsal KR. Radioimmunoassay monitoring of thyroid hormone concentrations in dogs on thyroid replacement therapy: 2,674 cases (1985-1987). J Am Vet Med Assoc 201: 623-629, 1992.

Nachreiner RF, Refsal KR, Davis WR, et al. Pharmacokinetics of L-thyroxine after its oral administration in dogs. Am J Vet Res 54: 2091-2098, 1993.

Nachreiner RF, Refsal KR, Graham PA, et al. Prevalence of autoantibodies to thyroglobulin in dogs with nonthyroidal illness. Am J Vet Res 59:951-955, 1998.

Panciera DL, Johnson GS. Hypothyroidism and von Willebrand factor. J Am Vet Med Assoc 206: 595-596, 1995.

Panciera DL. Clinical manifestations of canine hypothyroidism. Vet Med 92: 44-49, 1997.

Panciera DL. Thyroid-function testing: Is the future here? Vet Med 92: 50-57, 1997.

Panciera DL. Treating hypothyroidism. Vet Med 92: 58-68, 1997.

Panciera DL. Hypothyroidism in dogs: 66 cases (1987-1992). J Am Vet Med Assoc 204: 761-767, 1994.

Paradis M, Pagé N, Larivière N, et al. Serum-free thyroxine concentrations, measured by chemiluminescence assay before and after thyrotropin administration in healthy dogs, hypothyroid dogs, and euthyroid dogs with dermatopathies. Can Vet J 37: 289-294, 1996.

Paul MA. Credibility in the face of controversy. Am Anim Hosp Assoc Trends Magazine XIV(2):19-21, 1998.

Peterson ME, Melian C, Nichols R. Measurement of serum total thyroxine, triiodothyronine, free thyroxine, and thyrotropin concentrations for diagnosis of hypothyroidism in dogs. J Am Vet Med Assoc 211:1396-1402, 1997.

Schmidt MA, Bland JS. Thyroid gland as sentinel: Interface between internal and external environment. Altern Ther 3: 78-81, 1997.

Schultz RD. Canine vaccines and immunity: Important considerations in the success of a vaccination program, in Small Animal Immunology: New Faces of Immune-Mediated Diseases and Current Concepts in Vaccine Immunology. Proc San Diego Spring Vet Conf 1995;pp 100-113 .

Schultz RD. Current and future canine and feline vaccination programs. Vet Med 93:233-254, 1998.

Scott-Moncrieff JCR, Nelson RW. Change in serum thyroid stimulating hormone concentration in response to administration of thyrotropin-releasing hormone to healthy dogs, hypothyroid dogs, and euthyroid dogs with concurrent disease. J Am Vet Med Assoc 213:1435-1438, 1998.

Scott-Moncrieff JCR, Nelson RW, Bruner JM, et al. Comparison of thyroid-stimulating hormone in healthy dogs, hypothyroid dogs, and euthyroid dogs with concurrent disease. J Am Vet Med Assoc 212:387-391, 1998.

Smith CA. Are we vaccinating too much? J Am Vet Med Assoc 207:421-425, 1995.

Surks MI, Sievert R. Drugs and thyroid function. N Eng J Med 333: 1688-1694, 1995.

Thacker EL, Refsal KR, Bull RW. Prevalence of autoantibodies to thyroglobulin, thyroxine, or triiodothronine and relationship of autoantibodies and serum concentration of iodothyronines in dogs. Am J Vet Res 53: 449-453, 1992.

Thacker EL, Davis JM, Refsal KR, et al. Isolation of thyroid peroxidase and lack of antibodies to the enzyme in dogs with autoimmune thyroid disease. Am J Vet Res 56: 34-38, 1995.

Tizard I, Ni Y. Use of serologic testing to assess immune status of companion animals. J Am Vet Med Assoc 213: 54-60, 1998.

Tomer Y, Davies TF. Infection, thyroid disease, and autoimmunity. End Rev 14: 107-120, 1993.

Twark L, Dodds WJ. Clinical application of serum parvovirus and distemper virus antibody titers for determining revaccination strategies in healthy dogs. J Am Vet Med Assoc (in press, 2000).

Vaccine Technology in the 21st Century. Comp Cont Ed Pract Vet, Suppl 20: 1-39, 1998.

Vajner L. Lymphocytic thyroiditis in beagle dogs in a breeding colony: findings of serum autoantibodies. Vet Med Czech 11:333-338, 1997.

Williams DA, Scott-Moncrieff C, Bruner J, et al. Validation of an immunoassay for canine thyroid-stimulating hormone and changes in serum concentration following induction of hypothyroidism in dogs. J Am Vet Med Assoc 209: 1730-1732, 1996.

Table 1. Clinical Signs of Canine Hypothyroidism

Alterations in Cellular Metabolism

- lethargy
- mental dullness
- exercise intolerance
- neurologic signs
  - polyneuropathy
  - seizures
- weight gain
- cold intolerance
- mood swings
- hyperexcitability
- stunted growth
- chronic infections

Neuromuscular Problems

- weakness
- stiffness
- laryngeal paralysis
- facial paralysis
- "tragic" expression
- incontinence
- knuckling or dragging feet
- muscle wasting
- megaesophagus
- head tilt
- drooping eyelids
- ruptured cruciate ligament

Dermatologic Diseases

- dry, scaly skin and dandruff
- coarse, dull coat
- "rat tail"; "puppy coat"
- hyperpigmentation
- pyoderma or skin infections
- chronic offensive skin odor
- bilaterally symmetrical hair loss
- seborrhea with greasy skin
- seborrhea with dry skin
- myxedema

Reproductive Disorders

- infertility
- lack of libido
- testicular atrophy
- hypospermia
- aspermia
- prolonged interestrus interval
- absence of heat cycles
- silent heats
- pseudopregnancy
- weak, dying or stillborn pups

Cardiac Abnormalities

- slow heart rate (bradycardia)
- cardiac arrhythmias
- cardiomyopathy

Gastrointestinal Disorders

- constipation
- diarrhea
- vomiting

Hematologic Disorders

- bleeding
- bone marrow failure

Table 1 (continued)

low { red blood cells (anemia); white blood cells; platelets }

Ocular Diseases

corneal lipid deposits	corneal ulceration
uveitis	keratoconjunctivitis sicca or "dry eye"
infections of eyelid glands (Meibomian gland)	Vogt-Koyanagi-Harada syndrome

Other Associated Disorders

IgA deficiency	loss of smell (dysosmia)
loss of taste	glycosuria
other endocrinopathies	chronic active hepatitis
adrenal	
pancreatic	
parathyroid	

Table 2. Diagnosis of Thyroid Disease

- Complete Basic Profile
  -- (T4, T3, FT4, FT3, T4AA, T3AA)
- Additional Tests
  -- (TSH, TgAA)

- **Older Tests (T4, T4 + T3)**
  Serum T4 and/or T3 alone are **not** reliable for diagnosis because:

  -- overdiagnose hypothyroidism
  -- underdiagnose hyperthyroidism
  -- fail to dectect early compensatory disease and thyroiditis
  -- influenced by nonthyroidal illness and certain drugs

Table 2 (continued)

- **Newer Tests**

  Free (Unbound) T4
  Less likely to be influenced by nonthyroidal illness or drugs

  Valid
  -- equilibrium dialysis
  -- solid-phase analog RIA
  -- chemiluminescence solid-phase
  Less reliable -- liquid-phase analog RIA

  Endogenous Canine TSH
  In primary hypothyroidism, as serum free T4 levels fall, pituitary output of TSH rises.

  -- elevated TSH usually indicates primary thyroid disease
  -- 20-38% discordancy observed between expected and actual findings
  -- published normal ranges may need revising upwards
  -- affected by concomitant chronic renal disease

  Canine TgAA
  Thyroglobulin autoantibodies are present in serum of cases with lymphocytic thyroiditis.

  -- positive results confirm diagnosis
  -- 20% of cases have circulating T3 and/or T4AA
  -- allows for early diagnosis and genetic counselling
  -- needs validation in many breeds under field conditions

Table 3. Summary of Cases with Autoimmune Thyroiditis
January 1995 - January 1999

Total # of Dogs	Breed	Sex M; F; Unk	Mean Age	Mean T3AA	Mean T4AA	Rank Popularity 1995	1996	1997	1998
209	Golden Retriever	102 ; 96 ;11	4.2	5.7	1.8	4	4	4	2
124	Shetland Sheepdog	64 ; 57; 3	4.6	6.6	1.8	13	14	15	15
68	Cocker Spaniel (Am)	28 ; 40	5.0	6.4	1.4	7	8	8	13
51	Boxer	26 ; 25	3.8	8.2	1.6	15	13	13	12
42	Doberman Pinscher	17 ; 25	3.4	5.3	1.7	18	20	22	22
40	Labrador Retriever	17 ; 22; 1	5.2	5.2	1.4	1	1	1	1
19	German Shepherd	5 ; 13 ; 1	5.0	5.4	1.9	3	3	3	3
15	Akita	7 ; 4 ; 4	2.8	7.9	1.7	35	35	34	36
14	Irish Setter	5 ; 9	3.0	7.5	1.9	56	57	59	60
13	English Setter	3 ; 8 ; 2	4.6	4.7	1.2	83	87	86	91
12	Old English Sheepdog	4 ; 5 ; 3	4.1	9.8	1.9	55	56	60	65
10	Collie	4 ; 6	2.8	3.3	1.3	29	29	30	31
9	Poodle	6 ; 3	6.7	3.4	1.3	6	6	5	7
7	Skye Terrier	3 ; 4	4.9	2.7	1.1	133	135	130	130
7	Bull Mastiff	1 ; 4 ; 2	3.2	6.7	1.0	53	52	53	52
6	Scottish Terrier	4 ; 0 ; 1	1.7	9.8	1.6	41	41	43	42
6	Siberian Husky	1 ; 5	7.0	7.6	1.6	17	17	16	18
43	Mixed Breeds	18 ; 25	5.2	5.8	2.2				
269	Purebreds (< 5 dogs)	109 ;150; 10	4.7	4.7	1.5				
96	Unknown Breed	58 ; 38	3.8	7.5	1.6				
1060	Total Number of Cases		4.4	6.0	1.6				

Table 4. Canine Aberrant Behavior *

Total No. Cases	Purebreds	Mixed Breeds	Thyroid Dysfunction	Euthyroid
634	568	66	401 †	233

* Mean Age, 3.7 years (Range 0.5-12 years). Median Age, 2.5 years.

Table 5. Most Commonly Represented Breeds with Thyroid Dysfunction and Aberrant Behavior *

Breed†	Thyroid Dysfunction 401/634 (63%)	Aggression 251/634 (40%)	Seizures 189/634 (30%)	Fearful 55/634 (9%)	Hyperactive 42/634 (7%)
Golden Retriever	50/73	12/16	22/30	4/6	1/6
German Shepherd	34/53	10/22	14/16	3/7	2/2
Akita	27/38	24/33	0/1	0	0/2
Labrador Retriever	8/30	6/11	12/16	2/15	0/3
Shetland Sheepdog	14/25	3/6	2/3	2/4	3/3
Collie	8/9	0	7/7	0	0
English Setter	4/6	1/1	0	1/3	1/2
Other Purebreds	217/334	89/135	72/93	10/15	5/16
Mixed Breed	39/66	11/27	16/23	4/5	1/8
Totals	401/634 (63%)	156/251 (62%)	145/189 (77%)	26/55 (47%)	13/42 (31%)

* Some dogs had more than 1 abnormal behavior. Numerator = Thyroid dysfunction Denominator = Aberrant behavior † Total 634 cases; 72 dogs breeds represented.

Table 6: Age and Titer Results for CPV and CDV

Age in Years	Inadequate* CPV Titers	Adequate** CPV Titers	Inadequate* CDV Titers	Adequate** CDV Titers
Unavailable	1	42	1	40
< 1	5	33	3	21
1	5	84	5	82
2	7	87	1	88
3	4	123	2	121
4	9	132	2	134
5	7	113	1	112
6	7	111	1	113
7	2	119	2	117
8	11	134	2	136
9	0	86	1	81
10	4	90	5	85
≥11 ***	9	216	7	216
Total	71/1441 (4.9%)	1370/1441 (95.1%)	33/1379 (2.4%)	1346/1379 (97.0%)

* IFA <1:5 ** IFA ≥1:5 *** oldest dog was 17 years of age (titers were adequate).

Table 7: Time Between Last Known Vaccination and Antibody Titers of ≥1:5

Interval Between Last Vaccine and Titer (Years)	# of dogs for CPV	# of dogs for CDV
5	5	5
≥3, <5	44	41
≥2, <3	84	81
≥1, <2	268	263
<1 *	43	43
Totals	444	433

* 7 dogs were < 6 months old.

Veterinary Practice
STATE

Inherited and Acquired von Willebrand's Disease, Part 1

W. Jean Dodds, DVM
Sharon L. Raymond, BS
Marjory B. Brooks, DVM, Diplomate ACVIM
Hemopet
Santa Monica, California 90403

In humans and dogs, von Willebrand's disease (vWD) can be an inherited, congenital disorder present at birth and/or an acquired syndrome secondary to familial autoimmune thyroid disease.[1-7] In humans, acquired vWD also develops occasionally in association with cancer, systemic autoimmune disorders, and other causes.[3,4] Of the 57 dog breeds recognized to date to have vWD, 44 of them are affected with thyroid disease which usually progresses to hypothyroidism if left untreated (Figs. 1-3).[2,3,6-8] The prevalence of both

FIG. 1 — Roland, male Doberman Pinscher clinically affected with type I von Willebrand's disease (vWF:Ag 14%).

Bulk Rate
U.S. Postage
PAID
Permit No. 38
Fulton, MO

see von Willebrand's Disease, page 14

diseases has increased rapidly over the last decade despite the collective efforts of conscientious breeders working with veterinarians to test and screen out carriers and affected stock from their breeding programs.[6,7,9]

Genetic and Clinical Expression

INHERITED (CONGENITAL) vWD

An autosomal trait with variable clinical and genetic expression,[10] vWD is the most common mild inherited bleeding disorder of humans and animals.[2,7] In most affected purebreds or cross-breeds, vWD is classified as type I and is an autosomal incompletely dominant trait, which means it has variable expression within affected families.[5-7] The bleeding tendency can be manifested either by homozygotes that have inherited the gene from both parents or by heterozygotes that received the gene from either parent. Homozygosity is rarely seen in type I vWD, however, because affected puppies usually die during fetal development or shortly after birth.[1]

There is also an autosomal recessive form of vWD (type III) in which clinically affected individuals are homozygous for the vWD gene and have two asymptomatic, heterozygous (carrier) parents. Dogs affected with type III vWD produce virtually no von Willebrand factor (vWF) protein. The recessive disease has been recognized in Poland-China swine, Scottish Terriers, Chesapeake Bay Retrievers, Shetland Sheepdogs, Drahthaars (German Wirehaired Pointers), and several individuals of other breeds.[1,2,6,10] In some cases, animals clinically affected with type III vWD are actually the double heterozygous offspring of parents genotypically heterozygous for type I vWD (e.g., Shetland Sheepdogs).[6]

A rare type of vWD (type II) has been discovered in German Shorthaired Pointers and a family of Quarter Horses.[10,11] In this form of vWD, affected individuals produce an abnormal, dysfunctional vWF protein.

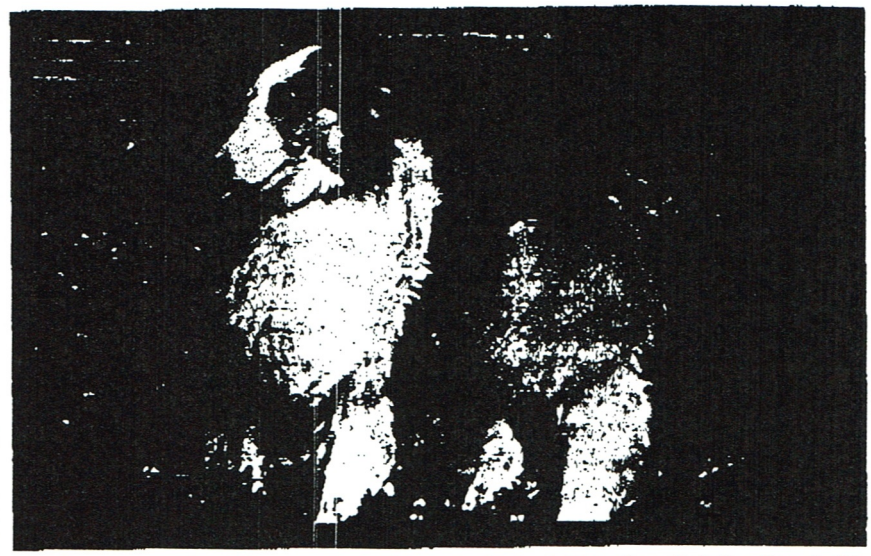


FIG. 2 – Amy and Strut, female and male littermate Shetland Sheepdogs clinically affected with type III von Willebrand's disease (vWF:Ag virtually absent). Both parents were heterozygous for type I vWD.

FIG. 3 – Topsy, female Golden Retriever clinically affected with type I von Willebrand's disease (vWF:Ag 42%). Despite excellent bloodlines, she had entropion as a puppy, developed radiographic changes of hip dysplasia, and became hypothyroid in mid-life. Hypothyroidism aggravated the clinical expression of her congenital vWD.

The type I disease is much more common than the other types of vWD and has been recognized in more than 50 breeds to date (Table 1).[7] Breeds with a high prevalence of the vWD gene include the Doberman Pinscher (80%), Scottish Terrier, Miniature Poodle, Pembroke Welsh Corgi, German Shepherd, Rottweiler, Standard and Toy Manchester Terrier, Keeshond, and the Standard and Miniature Dachshund (15-45%).[2] A severe form of vWD has been documented in a Himalayan and a Siamese cat.[2,12]

Although relatively few have severe problems and even fewer die from it, many animals are affected by vWD. Bleeding typically involves mucosal surfaces.[5,7,10] The bleeding episodes are worsened by physical, emotional, and physiological stresses as well as by other concomitant diseases. Typical

Continued

clinical signs include: recurrent gastrointestinal hemorrhage with or without diarrhea; recurrent hematuria; epistaxis; bleeding from the gums, vagina, or penis; lameness that mimics eosinophilic panosteitis; stillbirths or neonatal deaths ("fading pups") with evidence of bleeding at necropsy; prolonged bleeding at estrus or after whelping; bruises or hematomas on the surface of the body, limbs, or head; excessive umbilical cord bleeding at birth; and excessive bleeding from toenails cut too short or after tail docking, ear cropping, and dewclaw removal. Affected dogs may bleed to death from surgical procedures.[2,7,10]

ACQUIRED vWD

In dogs, vWD becomes more clinically severe if the dog also has hypothyroidism.[2,4,6-8] Thus, healthy carriers of the vWD gene may exhibit a bleeding tendency if they subsequently become hypothyroid, a situation commonly seen in many of the affected breeds. The relatively high incidence of both vWD and hypothyroidism combined with research on the synthesis and metabolic regulation of thyroid hormones and vWF have confirmed this link.[3,4,6-8] Hypothyroid dogs may also have thrombocytopenia which can contribute to mucosal surface bleeding. The bleeding tendency and laboratory abnormalities associated with hypothyroidism and vWD usually, but not always, resolve after treatment with appropriate doses of L-thyroxine.[2,4,8,13] It is generally impossible to distinguish between the inherited and acquired types of vWD in an individual patient with currently available techniques,[4,7] although it would be highly unlikely for a dog affected with acquired vWD to have zero or undetectable levels of von Willebrand factor antigen (vWF:Ag).[7,8]

In Doberman Pinschers, there is a very high prevalence of vWD along with an increased frequency and severity of bleeding episodes when thyroid disease is present, concurrently.[2,5-8,13,14] About 80% of the more than 23,000 Dobermans tested have < 50% vWF:Ag, and 18% of this total group exhibit a bleeding tendency.[2] Daily thyroid supplementation can lessen or control the clinical signs in dogs with both diseases and in some cases increases vWF:Ag levels two- or threefold.[2,4,8,13] Furthermore, circulating antithyroid antibodies are often present in humans and dogs with autoimmune thyroiditis (Hashimoto's disease) several years before the lymphocytic thyroid disease becomes clinically apparent. Animals with thyroid dysfunction can therefore have fluctuating levels of vWF and, when placed on thyroid supplementation, levels can increase to within normal limits. This could preclude the accurate diagnosis of their genetic status for vWD (i.e., carriers of vWD might test as normal when on thyroid medication). Because of the exacerbation of bleeding tendencies in dogs with vWD and hypothyroidism, breeders and veterinarians need to be aware of the increased risk and be more cautious about breeding or performing surgery on dogs with both problems.[7,8]

Diagnosis

The accurate diagnosis of vWD requires specialized vWF assays because the routine coagulation screening tests (activated partial thromboplastin time [APTT], prothrombin time [PT], and thrombin time) are nondiagnostic.[2] Affected individuals have long bleeding times, and definitive diagnosis is made by finding reduced or undetectable plasma concentrations of vWF:Ag and abnormal platelet-related assays of vWF.[5-7,9] Animals affected with the recessive type III disease are homozygotes having immeasurable vWF:Ag (0% vWF:Ag), whereas their heterozygous parents have reduced levels of vWF:Ag (< 50% of normal).[1,2,7] By contrast, animals affected with the incompletely dominant type I disease are heterozygotes having reduced but measurable vWF:Ag (1 to 49%).[7] Diagnosis of the rare type II

TABLE 1
Breeds of Dogs Known to be Affected With von Willebrand's Disease (As of March 1993)

A. Breeds With a High Prevalence of vWD Gene (15 to 80%)

Basset Hound	Toy Manchester Terrier
Miniature Dachshund	Miniature Schnauzer
Standard Dachshund	Pembroke Welsh Corgi
Doberman Pinscher (highest prevalence)	Miniature Poodle
German Shepherd	Standard Poodle
Golden Retriever	Rottweiler
Keeshond	Scottish Terrier
Standard Manchester Terrier	Shetland Sheepdog

B. Breeds With Either a Lower or Unknown* Prevalence of vWD Gene

Afghan Hound	Great Pyrenees
Airedale Terrier	Greyhound
Akita	Irish Setter
Alaskan Malamute	Irish Wolfhound
American Cocker Spaniel	Italian Greyhound
Australian Cattle Dog	Kuvasz
Azawakh	Labrador Retriever
Bearded Collie	Lakeland Terrier
Bichon Frise	Lhasa Apso
Boxer	Old English Sheepdog
Bulldog	Papillon
Cairn Terrier	Samoyed
Chesapeake Bay Retriever	Shih Tzu
Collie	Siberian Husky
Drahthaar (German Wirehaired Pointer)	Skye Terrier
English Cocker Spaniel	Soft-Coated Wheaten Terrier
English Setter	Swiss Mountain Dog
English Springer Spaniel	Tibetan Terrier
Smooth Fox Terrier	Vizsla
Wire Fox Terrier	Yorkshire Terrier
German Shorthaired Pointer	Whippet
Great Dane	

*Too few studied to date to estimate prevalence.

Continued

TABLE 2
Diagnosis of von Willebrand's Disease*

Bleeding Time**	von Willebrand Factor Antigen %+	Interpretation
N	70 or greater	Normal range is 70-180%
	70-79	Lower end of normal range; caution advised for breeding stock. Mates should have higher levels, and pups should be checked.
N	50-69	Borderline normal (equivocal result) or heterozygous carrier of vWD gene. Recommend retesting and/or breeding only to higher testing mates. Pups should be checked.
N	less than 50	Type I vWD. This is the most common form of vWD. Heterozygous carrier of vWD.* Recommend breeding only to higher testing mates. Pups should be checked.
Elevated	less than 50	Type I vWD. Has exhibited some bleeding problem (e.g., hematuria, epistaxis, melena, post-surgical bleeding).* Clinically affected animals should not be used for breeding.
N or elevated	less than 50	Type II vWD. Clinically affected animals exhibit severe bleeding symptoms. Multimeric analysis of vWF reveals absence of high molecular weight multimers. These animals should not be used for breeding. Has been reported in German Shorthair Pointers.
Elevated	less than 0.01	Type III vWD. Exhibits bleeding symptoms and is homozygous for vWD. This animal is the product of two asymptomatic, heterozygous carrier parents and should not be used for breeding.
N	greater than 180	Probably reflects stress, an improper sample, or activation from disease. Recommend retesting. Test invalid for prediction of genetic status for vWD trait.

+ Formerly called factor VIII-related antigen; measured by ELISA methodology. When measured by Laurell or RIA methods, normal ranges may vary slightly.
*Development of concomitant thyroid dysfunction may aggravate existing vWD or increase the risk of bleeding in some previously asymptomatic carriers.
**Abnormal bleeding time is not specific for vWD; hemostatic disorders including thrombocytopenia and platelet dysfunction will also cause variable prolongation of bleeding time tests.

Diagnosis of the rare type 2 form of vWD usually requires analysis of vWF multimer forms.[10,11] Table 2 summarizes the diagnostic findings in vWD.

vWF ASSAY

Assays of vWF protein are used clinically to diagnose individuals affected with vWD and as a means of identifying asymptomatic carriers of the vWD trait. The vWF:Ag assay is a quantitative measure of the protein's antigenic properties. This assay has been used routinely as a screening test for vWD.[9]

Until recently, most clinical laboratories used the Laurell electroimmunoassay methodology to measure vWF:Ag.[15] Alternative quantitative methods based on radioimmunoassay (RIA) and enzyme-linked immunosorbent assay (ELISA) techniques are now available.[14-18] The ELISA methodologies provide parallel accuracy to the Laurell method but are more sensitive (measure below 1% vWF:Ag vs. a lower detection limit of 7% by Laurell).[17] Additional benefits of the ELISA include increased efficiency and ease of standardization.

Our Albany, New York, laboratory switched over in 1990 to a newly developed ELISA system for measuring vWF:Ag.[16,17] We compared the vWF:Ag level determined by ELISA and Laurell techniques of over 600 randomly chosen, individual canine plasma samples submitted for testing.[16] We also looked closely at subsets of normal and affected dogs. In most individuals, the levels of vWF:Ag measured by ELISA were essentially the same as those determined by Laurell (correlation coefficient of 0.87). In a small number of cases (1 to 3%), the results of the two assays were discordant; that is, higher or lower vWF:Ag values were obtained on the same sample when measured with both techniques. These differences most likely reflect slight antigenic variations in part of the vWF molecule in these dogs, a situation also recognized in humans.[16] As more data are collected, including the results of pedigree analysis, vWF protein characterization, and thyroid testing, we hope to better understand the significance of these few discordant results.

A group of commercial veterinary testing laboratories now offer vWD testing. Most of them send their samples directly to our Albany, New York, laboratory, but a few do their own testing. Some human clinical laboratories also offer veterinary testing. It is advisable to be cautious here because routinely used human assays need to be adapted for use in animals to give meaningful results. A laboratory performing the test in-house should validate the assay for measuring canine vWF, check sample type and quality,

Continued

and note any unusual findings on the report. It is important to strictly standardize quality control and review procedures to minimize chances for an occasional error. The reproducibility of the ELISA vWF:Ag assay at our laboratory is ± 8% (intra- and inter-assay variation) with a coefficient of variation of less than 9%.[15,17]

Influence of Sample Collection, Processing, and Transport

Errors or difficulties encountered when collecting and processing blood samples for vWD screening still remain the most common reason for inconsistent results. Samples containing significant hemolysis or clots, even small ones, indicate that the animal was stressed and/or the sample was not obtained cleanly. Results for vWF:Ag on these samples may be artificially raised, lowered, or invalid.[15] One way to assure more consistent interpretation of vWD genotype for a breeding program is to retest all foundation stock if there is or was any question about the sample quality on an original test. Having more than one test result provides more confidence about the true reading of valuable foundation breeding stock.[9]

Progeny testing provides a means of verifying genotypic classification based on measurements of vWF:Ag. Results of vWF:Ag for both parents and entire litters should be provided for accurate interpretation. The expected outcome from matings of two parents genetically normal for the vWD trait is an entire litter of normal puppies. The presence of one or more carriers in such matings indicates misclassification of one or both parents.

When transporting samples for testing, they should stay cold or frozen in transit. An overnight mail or package express system is required. Samples should be placed in an insulated container with a frozen cold pack even in winter time because during transport the box could be kept indoors near a heating unit.[15]

Disparity Between Duplicate or Repeat Test Results

The most frequent cause of divergent results between so-called "duplicates" is that the specimens are not truly duplicates. Common mistakes in sending duplicates include taking separate samples from different veins one after the other (because the animal is more stressed for the second one) or dividing the plasma after centrifugation by removing the top portion, putting it in one tube and placing the bottom portion in a second tube. As plasma proteins centrifuge according to weight and mass, the heavier plasma proteins are at the bottom and lighter ones at the top. The correct way is to remove all the plasma, mix it thoroughly, and then divide it into two portions.

Another common problem arises when Vacutainers® (Becton Dickinson and Company, Sparks, Md.) are used to collect the blood sample. These tubes are designed for drawing the blood directly through the needle by vacuum into the tube. Many veterinary clinics, however, collect blood first into a dry, empty syringe and then add it to the tube containing the citrate anticoagulant. This modification allows for clotting to begin before anticoagulation occurs, and so samples are activated or partially clotted, which can produce inaccurate vWF:Ag readings.[15]

Production of vWF and Relationship to Autoimmune Thyroid Disease

The endothelial cells lining blood vessels are the primary and essentially sole source of vWF production in the dog and are the major source of vWF production in other species.[3,7,8,10] Any disease process that alters endothelial metabolism and protein synthesis can affect production and/or secretion of vWF.[8,13] Autoimmune diseases, especially thyroid disease, can disrupt vWF production and function resulting in the secondary, acquired form of vWD.[4,8,13] Therefore, lower levels of vWF are frequently, but not always, seen in dogs affected with or developing thyroid disease.

Blood Testing for vWD

The accuracy of the testing program in detecting the vWD genotype has been evaluated by retrospective analysis of the results to date for all three mating types (normal X normal; normal X carrier; and carrier X carrier).[9] The rate of misclassification of genetic status by this test was found to be less than 5%, which means that accurately measured levels of vWF:Ag are a reliable predictor of vWD genotype in healthy animals.[6,9] However, misclassification of genotype either way (false positive or false negative) is always possible. Some of the reasons for misclassification are:

- the animal belongs to the 5% not identifiable by this test;
- some other health problem such as hypothyroidism has altered vWF: Ag levels or the animal is receiving thyroid supplementation;
- the animal was in heat, pregnant, or lactating when tested;
- one or both parents have been incorrectly identified as being the true parent or as having a normal vWD genotype; or
- a collection, processing, or laboratory error was made. This last reason accounts for most of the misclassifications involving any type of genetic screening program.[9]

If a breeder plans a mating with another person's stud dog or brood bitch, wishes to lease or buy a tested animal, or wishes to buy a puppy of vWD-tested parents, it is prudent to advise them to see a copy of the results of vWF testing indicating the test status of the dog(s) beforehand, rather than rely on the memory or word of the owner. This is an added precaution and should not imply dishonesty or mistrust. ■

The second part of this article will be published in a future issue of the journal.

Inherited and Acquired von Willebrand's Disease, Part 2

W. Jean Dodds, DVM
Sharon L. Raymond, BS
Marjory B. Brooks, DVM, Diplomate ACVIM
Hemopet, 938 Stanford Street
Santa Monica, California 90403

Test Results and Interpretation

Results of our Comparative Hematology Laboratory von Willebrand's Disease (vWD) testing program are reported as the percentage of von Willebrand factor antigen (vWF:Ag) in comparison to an established pooled plasma standard from normal, healthy animals of the species being tested. The ranges for measurement of vWF:Ag utilizing the enzyme-linked immunosorbent assay (ELISA) technique are basically the same as those previously established for animals with the Laurell technique (Table 1).[15-17]

Dogs testing in the normal range are at low risk of transmitting the vWD trait. Dogs that are free of the vWD trait, when bred to a normal testing mate, should only produce offspring of normal genotype (i.e., with vWF:Ag levels > 70%).

When using the vWF:Ag test as a genetic predictor of vWD status, certain restrictions need to be applied before one considers test results falling within the normal range as valid. Measurements of vWF:Ag performed on animals that are ill or vaccinated within 10 to 14 days or on females during heat cycle, pregnancy, and nursing may be transiently higher or lower than the true baseline, and, therefore, may cause misclassification. Animals with endocrine disease, especially thyroid dysfunction, can also have fluctuating levels of vWF.[6-8,13]

There is an overlap region of vWF:Ag test levels (borderline range) between normal animals and those testing as carriers of vWD. Animals that test in this range cannot be accurately classified as free of the vWD trait or carriers on the basis of that measurement. We recommend that those testing in this range be bred only to mates testing well within the normal range. Their offspring should be tested to identify normal individuals and to further clarify the parent's vWD status. Healthy dogs testing in the abnormal range are considered to be carriers of the vWD trait and can transmit the defect to some of their offspring.[5,9]

"When using the vWF:Ag test as a genetic predictor..., certain restrictions need to be applied..."

We have seen a dramatic increase since 1985 in the number of purebred dogs having low levels of vWF:Ag (< 50%). When the data for Scottish Terriers and Shetland Sheepdogs were analyzed, a statistically significant increase in the prevalence of the vWD trait was found representing both inherited and acquired deficiency of vWF.[7] This overall increase is real because the number of dogs tested each year has continued to be large enough for statistical validation.[7] Several explanations could account for this increase. The first invokes a shift in the measurement of vWF:Ag; this possibility is ruled out because our assay has been standardized over time and so varies less than ± 8% of the actual value. Secondly, the prevalence of inherited vWD is increasing because tested or untested carriers continue to be bred as the popularity of these breeds is maintained. Finally, the increasing prevalence of thyroid disease has produced a parallel increase in acquired vWD. The most likely explanation is a combination of the last two possibilities.[7]

Management and Treatment

MANAGEMENT

Proper management and treatment of bleeding disorders cannot be achieved without an appropriate physiologic and physical environment for hemostasis, tissue repair, and prevention of recurrence. An extremely important aspect of medical management is the avoidance of the use of drugs known to interfere with hemostasis (e.g., aspirin, phenylbutazone, potentiated sulfonamides, estrogens, promazine derivatives, penicillin G).[1,2,4] These drugs are contraindicated for patients with moderate or severe hemostatic defects since they impair platelet function and further compromise the stability of the hemostatic plug.[2]

Continued

TABLE 1
Diagnosis of von Willebrand's Disease*

Bleeding Time**	von Willebrand Factor Antigen %+	Interpretation
N	70 or greater	Normal range is 70-180%
	70-79	Lower end of normal range; caution advised for breeding stock. Mates should have higher levels, and pups should be checked.
N	50-69	Borderline normal (equivocal result) or heterozygous carrier of vWD gene. Recommend retesting and/or breeding only to higher testing mates. Pups should be checked.
N	less than 50	Type I vWD. This is the most common form of vWD. Heterozygous carrier of vWD.* Recommend breeding only to higher testing mates. Pups should be checked.
Elevated	less than 50	Type I vWD. Has exhibited some bleeding problem (e.g., hematuria, epistaxis, melena, post-surgical bleeding).* Clinically affected animals should not be used for breeding.
N or elevated	less than 50	Type II vWD. Clinically affected animals exhibit severe bleeding symptoms. Multimeric analysis of vWF reveals absence of high molecular weight multimers. These animals should not be used for breeding. Has been reported in German Shorthair Pointers.
Elevated	less than 0.01	Type III vWD. Exhibits bleeding symptoms and is homozygous for vWD. This animal is the product of two asymptomatic, heterozygous carrier parents and should not be used for breeding.
N	greater than 180	Probably reflects stress, an improper sample, or activation from disease. Recommend retesting. Test invalid for prediction of genetic status for vWD trait.

+Formerly called factor VIII-related antigen; measured by ELISA methodology. When measured by Laurell or RIA methods, normal ranges may vary slightly.
*Development of concomitant thyroid dysfunction may aggravate existing vWD or increase the risk of bleeding in some previously asymptomatic carriers.
**Abnormal bleeding time is not specific for vWD; hemostatic disorders including thrombocytopenia and platelet dysfunction will also cause variable prolongation of bleeding time tests.

Live-virus vaccines and viral infections also affect platelet and/or endothelial function.[2-4] The effect occurs during the viremic phase after vaccination or exposure (usually at 5 to 10 days) and results in a relative thrombocytopenia, which may prolong the bleeding time and predispose the animal to hemorrhagic problems. Platelet reductions of 100,000/μl can occur. During this period, animals with hemostatic defects are at risk and should be watched for signs of bleeding. Elective surgical procedures such as ear cropping, spaying, castration, and dental surgery should be performed within 48 hours after vaccination or postponed for 10 to 14 days.[2]

TREATMENT

Until recently, the limited availability of blood products for routine use in clinical veterinary medicine has necessitated the development of alternate strategies for the treatment and management of bleeding disorders.[13,18] Table 2 outlines a stepwise approach for treatment of vWD. In those breeds in which both vWD and hypothyroidism occur relatively often, the administration of oral thyroid hormone (e.g., 0.1 mg/4.5 kg bodyweight given twice daily) has been effective in controlling bleeding episodes exacerbated by these concurrent diseases.[2,4,6-8,13] Clinical signs of bleeding are often lessened or controlled within 48 hours after therapy is initiated.[8,13] The therapeutic response should always be monitored with serial toenail or other bleeding times and periodically monitored by thyroid function tests. Dosages of thyroid medication should be adjusted accordingly to maintain thyroid levels of total T4, free T4, and total T3 within the upper half to third of the adult normal ranges.[4,13] In humans, L-thyroxine therapy has been shown to reduce levels of the circulating antibodies responsible for thyroiditis and hypothyroidism.[13] Presumably, this plays a role in the observed reversal of the bleeding tendency associated with thyroid disease. In dogs, the bleeding time is dramatically shortened and in some cases may even be corrected to within normal limits (< 5 minutes) from pretreatment values beyond 20 minutes.[13] Thus, thyroid supplementation alone may suffice to control bleeding in mild to moderate vWD, a situation analogous to the use of desmopressin (deamino-8-D-arginine vasopressin [DDAVP]) or danazol to control bleeding in humans.[4,10,13,14] DDAVP treatment has been shown to improve vWF levels in normal dogs and Doberman Pinschers with vWD,[14,18] although relatively high doses are required and the response is transient (1 to 3 hours).[4,13,18] Although most Doberman Pinschers benefit

Continued

TABLE 2
Treatment of von Willebrand's Disease

For short-term control of bleeding or prophylaxis for dogs at risk to bleed from vWD:

1. For elective surgery or invasive procedures, assess bleeding potential first with a toenail or buccal mucosal bleeding time. Normal values for dogs are 2 to 5 minutes.

2. Use L-thyroxine (T4) therapy at 0.1 mg/4.5 kg bodyweight twice daily for 7 to 10 days, if the bleeding time is prolonged or the patient is bleeding. Start 48 hours prior to elective surgery where applicable. Recheck bleeding time before performing procedure to ensure adequate hemostasis. Continue thyroid replacement beyond 10 days if patient is still bleeding or has thyroid disease. Thyroid supplementation is believed to promote hemostasis by improving platelet function, stimulating thrombopoiesis in bone marrow and other sites, and enhancing protein synthesis/release of vWF and other coagulation factors.[13]

3. An alternative drug for elective procedures is DDAVP (desmopressin) which causes a transient correction of the bleeding time in most dogs with vWD. Recommended dosage is 1 μg/kg given subcutaneously 20 minutes before anesthesia.[18]

4. Transfuse fresh-frozen plasma at 6 to 10 ml/kg of bodyweight once or twice daily, if bleeding continues or bleeding time is still prolonged after use of L-thyroxine. When available, cryoprecipitate* (cryoconcentrated plasma) can be used at doses equivalent to double those of whole plasma (12 to 20 ml/kg body weight) because an average 50% loss of activity occurs during preparation. For elective procedures (e.g., surgery on Doberman Pinschers with vWD), transfuse first and then perform surgery within 4 hours, because this period provides the maximum correction of the bleeding time.

5. For patients with PCV at or below 15%, transfuse packed red blood cells in saline at 6 to 10 ml/kg given once or twice daily. The packed cells can also be mixed with fresh-frozen plasma and transfused as reconstituted whole blood.

6. Avoid drugs or biologics that can impair hemostasis and/or induce thrombocytopenia. These include:

trimethoprim-sulfonamides	modified live virus vaccines
ormetoprim-sulfonamide	(3-10 days afterwards)
aspirin	estrogens
promazine tranquilizers	heparin
phenylbutazone	warfarin

*For control of severe bleeding crises or for homozygous-affected vWD patients undergoing surgery. In most cases, bleeding in animals with vWD can be controlled or prevented by using L-thyroxine, DDAVP, and/or fresh-frozen plasma.

from DDAVP treatment, certain individuals fail to respond, and the effect of this drug in other affected breeds has not been determined.[18] Hormonal therapy with DDAVP or thyroxine cannot induce vWF production in homozygous, congenitally vWF-deficient dogs having Type III vWD (zero level of vWF:Ag) and may not be beneficial in severe Type II vWD.[13,18]

Transfusion of fresh-frozen plasma or plasma cryoprecipitate along with packed red cells, where indicated, remains the treatment of choice for control of severe bleeding crises in vWD, for homozygous-affected vWD patients undergoing surgery, and for dogs that fail to respond adequately to L-thyroxine or DDAVP therapy.[2,13,18] ■

REFERENCES

1. Dodds WJ: von Willebrand's Disease in Dogs. Mod Vet Pract 65:681-686, 1984.
2. Dodds WJ: Bleeding Disorders. In: Handbook of Small Animal Practice. Ed by RV Morgan. New York, Churchill Livingstone Inc., 1988, pp 773-786; ed 2, 1992, pp 765-777.
3. Dodds WJ: Contributions and Future Directions of Hemostasis Research. JAVMA 193:1157-1160, 1988.
4. Dodds WJ: Bleeding and Immune Diseases, Parts I and II, and Acquired von Willebrand's Disease. St. Louis:Proceedings of the 56th Meeting of the American Animal Hospital Association, 1989, pp 606-619.
5. Brooks M, Dodds WJ, Raymond SL, Catalfamo J: von Willebrand's Disease: Signs, Causes, Treatment. Dogs in Canada. October 1989, pp 74-77.
6. Raymond SL, Jones DW, Brooks MB, Dodds WJ: Clinical and Laboratory Features of a Severe Form of von Willebrand's Disease in Shetland Sheepdogs. JAVMA 197:1342-1346, 1990.
7. Brooks M, Dodds WJ, Raymond SL: Epidemiologic Features of von Willebrand's Disease in Doberman Pinschers, Scottish Terriers, and Shetland Sheepdogs, 260 Cases (1984-1988). JAVMA 200:1123-1127, 1992.
8. Avgeris S, Lothrop CD Jr, Donald TP: Plasma von Willebrand Factor Concentration and Thyroid Function in Dogs. JAVMA 196:921-924, 1990.
9. Dodds WJ, Moynihan AC, Fisher TM, Trauner DB: The Frequencies of Inherited Blood and Eye Diseases as Determined by Genetic Screening Programs. JAAHA 17:697-704, 1981.
10. Johnson GS, Turrentine MA, Kraus KH: Canine von Willebrand's Disease: A Heterogenous Group of Bleeding Disorders. Vet Clin North Am 18:195-229, 1988.
11. Brooks M, Leith GS, Allen AK, Woods PR, et al: Bleeding Disorder (von Willebrand's Disease) in a Quarter Horse. JAVMA 198:114-116, 1991.
12. French TW, Fox LW, Randolph JF, Dodds WJ: A Bleeding Disorder (von Willebrand's Disease) in a Himalayan Cat. JAMA 190:437-439, 1987.
13. Dodds WJ: Blood Substitutes. In: Comparative Transfusion Medicine. Ed by SM Cotter. Adv Vet Sci Comp Med 36:257-290, 1991.
14. Meyers KM, Wardrop KF, Dodds WJ, Brassard J: Effect of Exercise, DDAVP, and Epinephrine on Factor VIII:C/von Willebrand Factor Deficient Doberman Pinscher Dogs. Thromb Res 57:97-108, 1990.
15. Benson RE, Jones DW, Dodds WJ: Efficiency and Precision of Electroimmunoassay for Canine Factor VIII-Related Antigen. Am J Vet Res 44:399-403, 1983.
16. Benson RE, Catalfamo JL, Brooks M, Dodds WJ: A Sensitive Immunoassay for von Willebrand Factor. J Immunoassay 12:371-390, 1991.
17. Benson RE, Catalfamo JL, Dodds WJ: A Multi-Species Enzyme-Linked Immunosorbent Assay for von Willebrand Factor. J Lab Clin Med 119: 420-427, 1992.
18. Kraus KH, Johnson GS: von Willebrand's Disease in Dogs. In: Current Veterinary Therapy X, Small Animal Practice. Ed by RW Kirk. Philadelphia, WB Saunders, Inc, 1989, pp 446-451.

Part 1 of this article was published in the July/August 1993 issue of *Veterinary Practice STAFF.*